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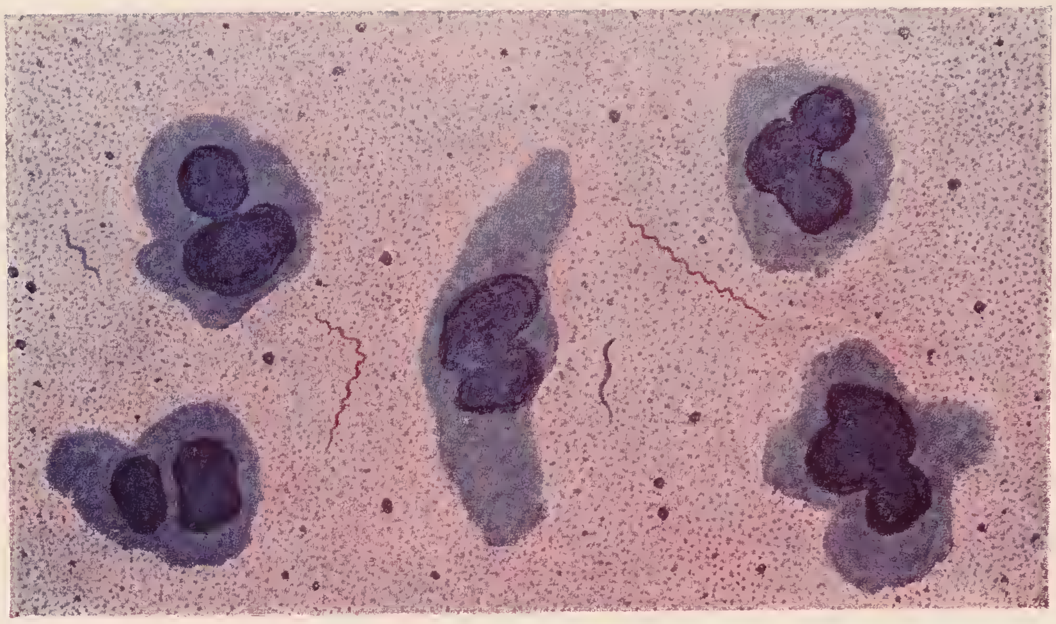

FIG. A.—Spirochæta pallida and Spirochæta refringens. Smear from a primary syphilitic sore, fixed in osmic vapour and stained by Giemsa's method (leucocytes necessarily overstained).

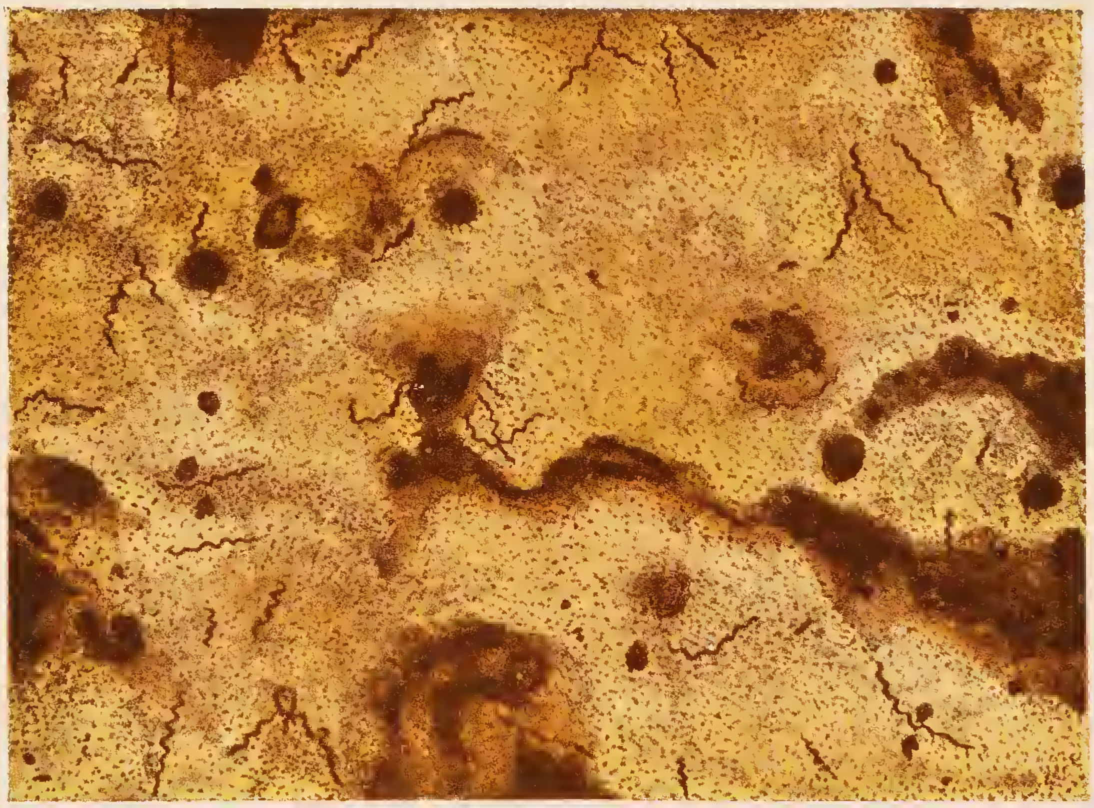

FIG. B.—Spirochæta pallida in liver of syphilitic fœtus. Stained by Levaditi's silver method.

SPIROCHÆTES

A REVIEW OF RECENT WORK
WITH SOME ORIGINAL OBSERVATIONS

BY

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PREFACE

HAVING had occasion to study the literature bearing on the subject of Spirochætes in connection with observations which I made on two species of this genus, I was struck with the voluminous and at the same time scattered mass of papers to which reference had to be made. I therefore thought it might be useful to others if, instead of merely laying aside the abstracts which I had made, I put them together in the form of a small book.

The spirochætes present a problem of considerable interest. Their position in the scale of living things is still undecided, and there is a tendency to place them in a position between the bacteria and the protozoa. My own observations would lead me to regard them as much more closely allied to the former than to the latter.

I have endeavored, in the first part of this work, to compare the statements of different writers on the morphology and behavior of spirochætes, with a view to establishing, if possible, the features characteristic of the genus. In the second part, I have dealt with the separate species described. I have confined myself to very brief descriptions of the various organisms and of the methods employed for examining them, as those who are interested in particular species or methods will in any case refer to the original papers. Their task will, I hope, be facilitated by the bibliography here appended. This is by no means exhaustive, as many papers are published in unattainable periodicals, and I

have included comparatively few references to articles which I have not myself consulted either in the original or in abstract. I hope, however, that the more important papers are included.

My thanks are due to Mr. A. N. Leathem for the loan of specimens, and to Professor E. A. Minchin for the very kind loan of books from his library and of specimens in his possession, as well as for permission to use his laboratory for my own small researches.

LONDON, ENGLAND,
May, 1911.

W. C. B.

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SPIROCHÆTES.

A REVIEW OF RECENT WORK.

SECTION I.

GENERAL CHARACTERS OF SPIROCHÆTES.

BIOLOGICAL POSITION OF SPIROCHÆTES.

The name Spirochæta was first invented by Ehrenberg in 1833 for the organism discovered by him and named *Spirochæta plicatilis*. It was regarded by him as a protozoon, but its relationship to other members of this order was not clearly determined; and while some writers considered that spirochætes were members of the class Trypanosomidæ, others regarded them as really bacteria. Indeed, in some of the earlier textbooks of bacteriology the term spirochæte was applied to a number of spirillar forms.

The great increase of interest which has recently been aroused in the spirochætes dates from the discovery by Schaudinn in 1905 of the spiral organism associated with syphilis, which he assigned to this class. The attention thus directed to these minute organisms has resulted in the discovery of a large number of forms, some associated with conditions of disease, others living apparently as harmless saprophytes. Among the latter must be classed the large varieties present in several genera of shell-fish.

The relationship existing between the larger and the smaller spirochætes has not been thoroughly eluci-

dated. Indeed, owing to the minute size and feeble staining properties of the latter organisms, it is difficult to recognise more than their shape and movements. Details of finer structure, which might throw light on their true nature, are as yet wanting. The type of the smaller spirochætes may be seen in the organism associated with the disease called relapsing fever, usually called *Spirillum* or *Spirochæta obermeieri*, after its discoverer, but perhaps more correctly on the basis of recent biological nomenclature, *Spirochæta recurrentis*, the spirochæte of recurrent fever. This organism was for many years regarded as a bacterium and placed in the same class as the spirillum or vibrio of cholera. It does not seem proved so far that this classification is incorrect, and the question is still at issue, names of weight being found on both sides in the controversy as to the protozoan or bacterial nature of the smaller spirochætes. Even the larger spirochætes have by some been assigned to the bacteria as, for example, by Schwellengrebel, who points out the close resemblance between an organism, such as *Sp. balbianii* and some of the larger spirilla, such as *Spirillum giganteum* (*Spm. volutans*). I hope to show in subsequent pages reasons for thinking that this view is probably correct.

With regard to the larger spirochætes, it is certainly natural to anyone who observes the extreme activity of movement exhibited by such an organism as *Sp. anodontæ*, to regard it as *prima facie* an animal. This vigour of movement, however, does not by itself constitute an argument of appreciable weight, for among the spirilla, which are generally admitted to be bacterial, very active movement may be observed. Nevertheless, there is some difference between the two classes in this respect; for while the spirochætes perform vigorous lashing movements in which the whole body is bent to-

and-fro like the lash of a whip, the spirilla confine themselves almost entirely to a rapid "cork-screw" motion, with little alteration in the long axis of the body. A certain degree of the latter motion may, however, at times be observed in them, so that this difference is rather quantitative than one of kind.

Failure to cultivate spirochætes, large or small, in artificial media has been urged as a reason for regarding them as protozoa and not bacteria, but this argument again is of little weight, since not all known bacteria can be grown artificially (*e.g.*, *B. lepræ*), and the quality of adaptability to cultivation in the laboratory can hardly be looked upon as essential to this or any other class. Indeed, some of the protozoa can be cultivated; for example, some forms of amœbæ and of flagellata.

The curious arrangement of the staining material in the body of the larger spirochætes, which has been regarded as constituting a special nucleus, has been held to prove their animal nature; but there is at present too much uncertainty as to the constitution of this staining substance and its formation in these organisms to allow of any great weight being assigned to this peculiarity for purposes of classification. Schwellengrebel states that a spiral filament similar to that found in *Sp. balbianii* may be found in *Spirillum giganteum*, though his statement has not so far been confirmed. Certainly a resemblance exists between the form taken by the staining substance in this spirillum and that seen in some spirochætes (Figs. 8, 9, 10).

Other peculiarities of the large spirochætes which are regarded as protozoan characteristics are the asserted existence of an undulating membrane, the lack of flagella, and the habit of longitudinal division for purposes of multiplication. With regard to the first of

these, doubt has recently been thrown on the identity of the appearance seen in spirochætes with the true undulating membrane of such organisms as trypanosomes. Consequently it is premature to regard this as a distinctive feature. Flagella have been described in some of the smaller spirochætes, but with doubtful certitude; they appear to be absent in the larger forms. Such absence does not seem to constitute an argument for or against their vegetable nature. The method of reproduction even in the larger spirochætes is still disputed, but there appears to be a balance of authority in favour of longitudinal division as the usual occurence, and I have myself seen an instance of what appeared to be this procedure in *Sp. anodontæ.*[1] The occurrence of encysted forms in such species as *Sp. balbianii* has also been held to prove them to be protozoa, but the real nature of the curled up forms is as yet uncertain, and it is not unknown for vegetable organisms to enter upon a resting stage.

The strongest argument which could be brought forward in favour of the animal nature of the spirochætes would be the occurrence of conjugation—a distinctive protozoan characteristic. This phenomenon has been described as occurring in several species of spirochætes (see p. 37), but the evidence adduced is unconvincing.

The nature of the smaller spirochætes is still more uncertain. The points about them which seem to place them in the same class as the larger spirochætes are their general shape and movements, the occurrence of longitudinal division, the asserted possession by some of them (not by all) of an undulating membrane, and the absence of flagella. Their resemblance to the larger forms in the matter of general shape and movements is

[1] If Schmeidlechner's observation of bacilli which divide longitudinally is confirmed, this mode of multiplication cannot be considered a distinguishing feature of animal organisms.

certainly striking, but is not in itself a strong argument for combining them in a single group. The possession of an undulating membrane by these minute organisms is certainly doubtful, and the presence or absence of flagella has little weight in this connection, except as an arbitrary ground of classification. The question as to the multiplication of these little organisms by longitudinal or transverse division is still undecided; but the formation of a fine thread at the point of division seems to be established, whereas bacteria divide by means of a distinct septum. The argument from the possibility of cultivation on artificial media has just been noticed to be valueless, and indeed some of the small spirochætes have, if the claims of various writers are to be believed, been successfully grown in the laboratory. The conveyance of some of the pathogenic spirochætes by biting insects (ticks) has been used as a proof of their animal nature, but can hardly be regarded as a conclusive argument; for it has not been proved that they undergo any constant process of development within these hosts.

A stronger argument has been drawn from the action of certain chemical reagents on small spirochætes. Thus it is said that cobra-venom, taurocholate of sodium, saponin, and pancreatic extract destroy spirochætes and protozoa, but do not affect bacteria. Spirochætes are little affected, however, by distilled water or by concentrated salt-solution. It is also asserted that whereas bacteriolysins are thermostable (not destroyed by heat), the corresponding substances which act on spirilla (spirochætes) are thermolabile, thus resembling the hæmolysins. On the other hand, the action of immune serum in producing agglomeration or agglutination of spirochætes is like that exerted upon bacterial organisms; and *Sp. pallida*—if this be really the cause

of syphilis—gives rise in the body of the human host to the formation of an antibody (copula or amboceptor) as do the pathogenic bacteria. This form of immune substance has not been found in infections caused by protozoa.

It is to be hoped that further study of the life-history and development of spirochætes will throw definite light on their true biological position. If the observation of Leishman on the development of *Sp. duttoni* into coccoid forms be confirmed, and if a similar change can be shown to take place in other spirochætes, as is suggested by my own experience, their close relationship to the bacteria will be manifest.[1]

In attempting to assign the spirochætes either to the protozoa or the bacteria, it must be borne in mind that there are several other kinds of organisms known, about which there is equal uncertainty as to their correct classification. Indeed, it has to be confessed that there are no definite criteria, by which to draw a line of division between the lowest forms of animals and plants. This must be regarded as almost inevitable, since both animal and vegetable kingdoms present to us as their lowest members unicellular organisms of very simple structure, while it is at least probable that at the beginning of things a single form of living matter gave origin to both the great divisions, animals and plants.

MORPHOLOGY OF SPIROCHÆTES.

The minute structure of the **larger spirochætes** has been carefully observed in the species *Sp. balbianii* and

[1] The suggestion that spirochætes are stages in the development of a bacterial organism is referred to on page 42. Schaudinn's original view that they constitute a stage in the life-history of a trypanosome is now generally supposed to have been based on a mistaken identification. (See Novy and MacNeal, *Journ. Infect. Dis.*, 1905, II, 256; Sergent, Edm. and M., *Ann. d. l'Inst. Pasteur*, 1907.)

Sp. anodontæ, which are easily obtainable, as well as, to a less extent, in the typical species *Sp. plicatilis*. De-

FIG. 1. FIG. 2. FIG. 3.

FIG. 1.—Spirochæta plicatilis: Schematic. (After Doflein.)
FIG. 2.—Spirochæta balbianii, showing so-called "undulating membrane." (Perrin.)
FIG. 3.—Spirochæta anodontæ, with "undulating membrane." (Keysselitz.)

scribed in general terms, they are elongated, vermiform organisms, endowed with active movement. They are said to be flattened from above downward, thus

resembling strips of ribbon rather than threads of cotton in form. They are further stated to be furnished with an undulating membrane and with a complicated nuclear apparatus (see Figs, 1, 2, 3).

Shape.—The length of these organisms is usually not less than twenty times the breadth, *Sp. balbianii* for example reaching a length of 100μ, while its breadth is usually less than 5μ. The length of *Sp. plicatilis* may reach 500μ. I have found specimens of *Sp. anodontæ* nearly 130μ in length, without any signs of division at any point; but unusually long forms are regarded by some writers as about to enter upon the process of fission.

It is difficult to ascertain whether these organisms are naturally flattened in form, as during active movement, while they are alive, the exact contour cannot be made out, while some flattening might easily take place after death in the process of fixing. *Sp. balbianii* is said by Perrin to be round in section. Fantham and Gross have succeeded in obtaining transverse sections of spirochætes, which suggest a form such as that shown diagrammatically in figure 4—a rounded organism with a loose sheath which may project on one side. The organism might thus appear flattened in one direction, and round in another; and the opposing views might thus be harmonised. Gross regards the projecting portion as constituting a crest or comb.

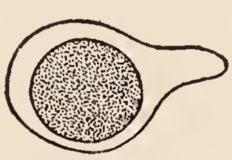


FIG. 4.—Diagram of spirochæte in transverse section.

The ends of the organism may be either blunt or pointed (Figs. 2, 3). It is uncertain whether this feature is always constant in the same species. Thus, in the case of *Sp. anodontæ* Schellack would constitute the individuals with pointed ends[1] a separate species (*Sp.*

[1] Cf. Castellani's distinction of *Sp. obtusa* and *Sp. acuminata*, page 113.

spiculifera). Certainly this form tends to be shorter on the average than the form with blunt extremities; but some overlapping may occur, and it seems premature to make a specific division on this ground alone (see page 81).

Movements.—The movements of the large spirochætes are very rapid under normal conditions, but become slower as vitality diminishes. They are of three main kinds: (1). Lashing movements, such as are performed by an eel suspended on a hook, the whole body being bent from side to side, taking the forms of a circle or an S or a figure of eight. These are most evident when the spirochæte is attached in some way by one extremity or hindered in its progression. (2). Undulating movements, which are compared to the flapping of a sail "in the wind," a wave passing from one end of the organism to the other and being rapidly succeeded by other similar waves in the same direction. It would seem that the direction may be reversed. (3). A rotatory movement, like that of a cork-screw when pushed into a cork, the whole spirochæte turning on a longitudinal axis passing through the centres of the spirals (the axis of the helix). Special forms of movement are described by Perrin as taking place in the process of encystment, one end of the organism gliding down the side of the body, to be followed by a similar movement of the other extremity, and finally by a continuous movement by which the creature is rolled upon itself.

Structure.—Examined microscopically, the structure of the spirochæte is seen to consist of an external coat or *periplast*, which contains an inner protoplasmic substance or *entoplasm*. The periplast may be prolonged at one or both ends of the organism into an appendage, which some authors regard as a flagellum. This identification seems incorrect. It has also been suggested

that these appendages are formed in the process of division of the spirochæte, and thus may not be permanent. Perrin speaks of the periplast retaining its form after the entoplasm has escaped from within it and become encysted.

Attached to the periplast or forming part of it is the so-called *undulating membrane* (Figs. 2, 3). This is described as running round the body of the spirochæte in a spiral direction, and as being bounded at its external margin by a darkly staining fibril (*randfibrille*), while other fibrils may be seen running in it parallel to the marginal one. The latter is said to be connected at its extremities with two darkly staining particles, situated at the ends of the spirochæte, called by Gonder blepharoplasts, on the analogy of the blepharoplast of trypanosomes.

The nature of this undulating membrane has been called in question. Laveran and Mesnil regard the appearance thus named as produced by the existence of a loose sheath outside the spirochæte; and a sheath has been described in several of the smaller spirochætes by different writers (see p. 26). Schellack believes that the so-called undulating membrane is an artifact, produced in the process of fixation. My own experience with *Sp. anodontæ* would seem to support this view, as I find that the better the specimen is fixed, the less frequent are individuals which appear to have undulating membranes. Nor does the appearance when present correspond at all closely with that seen in trypanosomes, the marginal fibril being often seen as a separate structure with no membrane visible between it and the body of the spirochæte, and often also appearing at only one or two points, not as a structure continuous all along the organism. The appear-

ance of this fibrillary structure at the outer margins of the curls presented by the spirochæte suggests that it may be a portion of periplast which has become fixed to the slide, and has thus remained fast, in a figure corresponding with the curves in which the creature lay at the moment of death: then as drying took place and the body of the spirochæte shrank, the latter straightened out somewhat, while the adherent periplast remained. Sometimes, however, the "membrane" runs in a straight line between neighbouring coils, having apparently shrunk more than the body-substance.

Borrel and Cernovodeanu, however, state that they have seen an undulating membrane in living specimens of *Sp. anodontæ.*

Perrin thinks that the undulating membrane is not of much importance as an aid to locomotion in *Sp. balbianii.*

With regard to the structure of the entoplasm and its contents an equal degree of uncertainty prevails. Perrin described in *Sp. balbianii* the existence of peculiar form of *nucleus.* It consists, according to this writer of a spiral band of achromatic substance (karyosome) on which appear at intervals masses of staining material (chromatin?). These latter usually, in resting organisms, take the form of rods arranged so as to lie transversely, more or less at right angles to the main axis of the spirochæte (Fig. 5); but when fission is about to occur, the rods assume other forms and undergo division. Fantham confirmed Perrin's statements. Keysselitz described in *Sp. anodontæ* balls and rods of chromatic material (Fig. 6), and thinks that there may be a spiral arrangement such as Perrin found, but did not himself observe it. Schellack confirms the existence of bands of chromatin, but denies that they are connected

by any spiral thread. He regards the spirochæte as made up of a series of chambers, in which these rod-shaped nuclei lie, and thinks that the increase in length of the spirochæte is effected by a multiplication of these

Fig. 5.

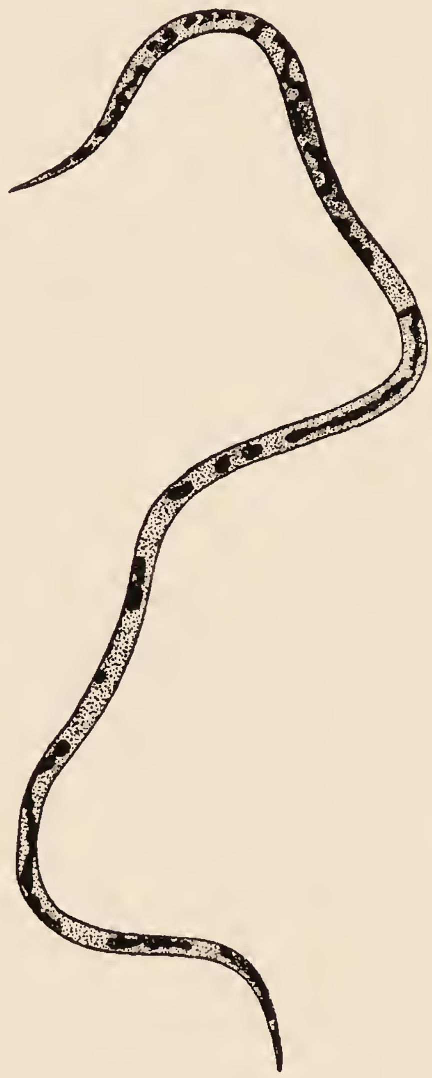


Fig. 6.

Fig. 5.—Spirochæta balbianii, showing nuclear (?) bands. (Perrin.)
Fig. 6.—Spirochæta anodontæ, with irregular masses of staining substance. (Keysselitz.)

chambers, taking place especially at the ends. In some cases the staining substance appears as a central continuous band or rod, running along the middle of the body of the spirochæte (Fig. 7).

The appearances seen in *Sp. plicatilis* are somewhat different (Fig. 1). Here the organism appears to be furnished with a series of rounded nuclei, distributed

Fig. 7.—Spirochæta balbianii; rod-shaped "nucleus." (Perrin.)

fairly evenly throughout its length. The general outline of the spirochæte is also different, since it shows not only the large undulations seen in *Sp. anodontæ* and *balbianii*, but also a series of smaller waves superimposed upon the others. It has no undulating mem-

Fig. 8.—Spirochæta anodontæ, showing irregular granules of staining material.

brane, as depicted in Doflein's illustration—indeed, judged merely by a comparison of this with the illustrations of *Sp. balbianii*, given by Perrin or Fantham, it

is difficult to believe that the two organisms belong to the same genus.

So far as I can judge from my own observations, the peculiar aggregations of staining material seen in *Sp.*

Fig. 9.

Fig. 10.

Fig. 9.—Bacteria from rectum of ox for comparison with Fig. 8.
Fig. 10.—Spirillum volutans for comparison with Fig. 8.

anodontæ are not to be looked on as nuclei. They are irregular in shape and quite unlike the nuclei of protozoa. They seem to occur for the most part in organisms which are degenerating (so-called "involution

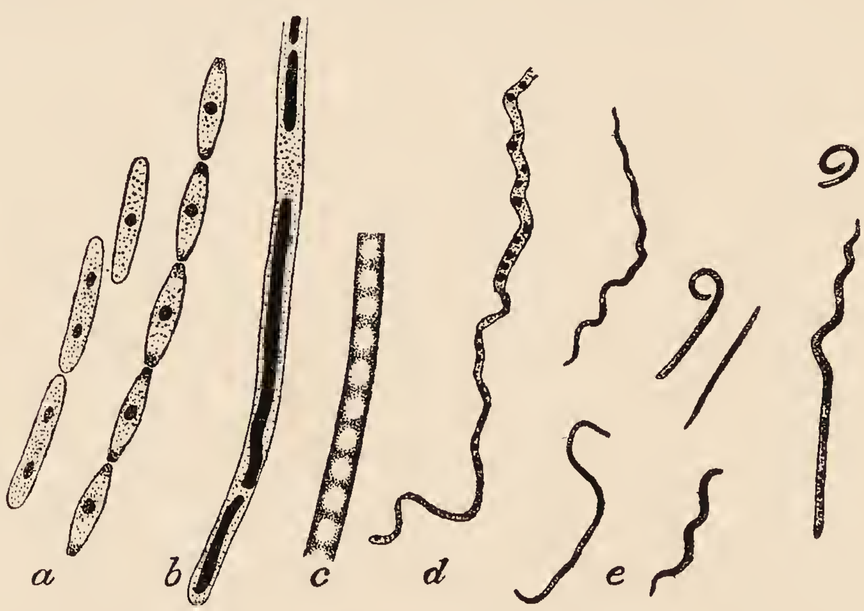


Fig. 11.—Filaments of algæ: *a*, Resembling fusiform bacilli; *b*, showing central staining rod; *c*, with series of chambers; *d*, *e*, curved like spirochætes.

forms"). In other instances they may point to a forthcoming developmental change (p. 40). The great majority of the organisms in my experience stain

homogeneously like bacteria. In some a vacuolated structure may be made out, resembling that seen in some of the larger spirilla. In the larger bacteria also granules or masses of staining material may be seen, not unlike those in *Sp. anodontæ* (see Figs. 8, 9, 10). Filaments of algæ may also show features bearing a close resemblance to some of those described in spirochætes (Fig. 11).

In the case of the **smaller spirochætes** the minute size of the organisms renders the investigation of their intimate structure almost impossible. The general shape is the same as that of the large varieties, according to most writers, but it is even more difficult to make sure of their flattened shape than in the larger organisms. Some are definitely stated to be round in section, *e.g.*, *Sp. pallida*, but many authors follow Vuillemin and Schaudinn in placing this and a few others in a separate genus, *Treponema*, partly on this account.

The *movements* of the small spirochætes are the same as those described above. An additional form of movement is, however, described by Plaut in *Sp. vincenti* and called by him "euglenoid" movement: it consists in the appearance of a thickening at one point in the body of the organism and the gradual passage of this swelling from one end to the other. Krzystalowicz and Siedlecki describe a somewhat similar compression and extension of the body of *Sp. pallida* when it is in motion. It must be remembered that in these minute thread-like organisms it is impossible to distinguish under the microscope between rotatory movement and undulation. Lashing movements are said to be less marked in treponema: indeed, the movements *Sp. pallida* (*Trep. pallidum*) are altogether sluggish.

Of the actual *structure* of these tiny organisms little can be said. An undulating membrane has been described in some species, as by Gonder in *Sp. vesperuginis*, by Prowazek in *Sp. gallinarum*, by Hoffmann in *Sp. buccalis*, and by Schaudinn in *Sp. refringens*. On the other hand, Levaditi denies that *Sp. refringens* possesses an undulating membrane, and this structure has never been seen or described in *Sp. obermeieri*, *Sp. pallida*, *Sp. muris*, the spirochæte of ulcerating cancer, etc. When it is borne in mind that one of the favourite stains for spirochætes, Giemsa's, is deposited round, as well as in, the structures which attract it, it is easy to imagine a

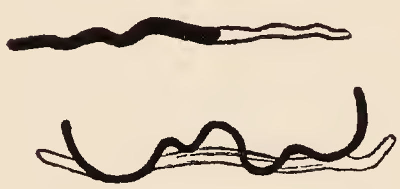


FIG. 12.

FIG. 13.

FIG. 14.

FIG. 12.—Spirochæta duttoni; appearance of a sheath. (Stephens.)
FIG. 13.—Spirochæta buccalis. Sheath. ×2250. (Prowazek.)
FIG. 14.—Anthrax bacillus, with appearance of a sheath. (Ascoli.)

possible source of error in observing this point in a minute organism of spiral shape. Most of the illustrations of undulating membranes in these small spirochætes are far from convincing.

A definite *sheath* has been described in some forms, *e.g.*, in *Sp. duttoni* by Stephens (Fig. 12), in *Sp. pallida* by Leuriaux and v. Geets, in *Sp. buccalis* by Prowazek (Fig. 13), in the spirochæte found by Baruchello and Pricolo in equine pneumonia, and in an unidentified spirochæte found by Kenrick in a case of fever. An appearance resembling a sheath, may be found in bacteria, as is shown in the accompanying illustration of

Bacillus anthracis (Fig. 14), taken from a paper by Ascoli.[1]

Terminal *flagella* are described in some forms, *e.g.*, *Sp. obermeieri* (Reichert), and *Sp. duttoni* (Stephens),

FIG. 15.—Spirochæta pallida, with terminal filament; so-called autogamy stage. (Prowazek.)

but these are generally regarded (as by Schaudinn and by Krzystalowicz and Siedlecki) as mere prolongations of the periplast (Figs. 15, 16, 17, 18). Lateral flagella have also been described (*e.g.*, by Fraenkel in *Sp.*

FIG. 16.

FIG. 17.

FIG. 18.

FIG. 19.

FIG. 16.—Spirochæta, media of stomatitis, with terminal filament. ×2250. (Prowazek.)
FIG. 17.—Spirochæta duttoni (terminal filament). (Schellack.)
FIG. 18.—Spirochæta balbianii, with terminal "brush." (Schellack.)
FIG. 19.—Spirochæta recurrentis; splitting of periplast. (Schellack.)

obermeieri), but are probably due to injury in fixation of specimens (Fig. 19).

Small *granules* or *nodules*, appearing rather larger in diameter than the rest of the spirochæte, are commonly

[1] *Centralbl. f. Bakt.*, 1908, XLVI, 186.

seen. A single terminal nodule has been noted, for example, in *Sp. duttoni* (Dutton, Todd and Tobey), *Sp. pallida* (Dudgeon), *Sp. pertenuis* (Castellani), and in

Fig. 20.

Fig. 21.

Fig. 20.—Spirochæte of ulcerating cancer (left) and Sp. refringens (right), with terminal nodules. (Schaudinn.)

Fig. 21.—Spirochæta pertenuis with terminal granules. (Castellani.)

many other forms (Figs. 20, 21, 22, 23). Two nodules may be placed toward the same extremity (*e.g.*, *Sp. aboriginalis*, Bosanquet). One or more of such dots or masses may also be seen (Figs. 24, 25), in the central

Fig. 22.

Fig. 23.

Fig. 24.

Fig. 25.

Fig. 22.—Spirochæta aboriginalis with granules and "loop." (Bosanquet.)

Fig. 23.—Spirochæta duttoni. Injured specimen. (Schellack.)

Fig. 24.—Spirochæta duttoni, with masses of staining substance. Supposed to represent nuclei. ×2200. (Mayer.)

Fig. 25.—Spirochæta lagopodis, showing dots resembling nuclei. (Fantham.)

part of the organism. These have been supposed to represent nuclei, and Herxheimer has indentified a kineto-nucleus, a tropho-nucleus and centrosome—a

degree of refinement which at present seems fanciful. These little nodular bodies sometimes appear to be attached to the side of the spirochæte rather than to lie in its body (Fig. 26). It is not unusual to find spirochætes which resemble a chain of granules set like a string of beads (Fig. 27): these are usually regarded as degenerated forms ("*moniliform degeneration*"), but the possibility that these organisms break up into

FIG. 26.

FIG. 27.

FIG. 26.—Spirochæta gallinarum, with lateral nodule. (Prowazek.)
FIG. 27.—Spirochæta obermeieri, showing breaking up of body into rods and granules (diagrammatic).

minute bodies capable of growing into other individuals, when placed in favourable circumstances, cannot be excluded.

A *loop* has been described as present at one end of the spirochæte in certain forms, *e.g.*, in *Sp. pallida* (Reuter), *Sp. pertenuis* (Castellani).

Masses of organisms may be found tangled together, as in *Sp. pallida* (Bandi and Simonelli), *Sp. dentium* (Miller), *Sp. obermeieri* (Zettnow), and *Sp. duttoni* (in the tick, Koch). I have once seen a large number of *Sp. anodontæ* forming a tangle.

VARIABILITY OF FORM.

A very important question is that of the possibility of some degree of change of form in a single species, since upon the negative reply to this question must at present depend the possibility of accurately distin-

guishing and classifying these organisms. The existence of somewhat different forms side by side has been already referred to in connection with *Sp. anodontæ*. It is remarkable that two forms also exist together in Tapes læta (*Sp. tapetos*) and in other molluscs (Schellack). The coexistence of the two forms, *Sp. pallida* and *Sp. refringens*, in syphilitic lesions is well known. Two forms are described by Loewenthal in ulcerated cancers, one larger and the other smaller, and two also by Branch in cases of hæmoptysis, and by Moritz in decaying potato (*Sp. polyspira*). Castellani found four forms associated in his cases of hæmoptysis.

The question of change of form has been most carefully studied in connection with *Sp. pallida*. Bertarelli and Volpino describe intermediate forms between *Sp. pallida* and *Sp. refringens*, and Bosc suggests that *refringens* is a degenerative form of *pallida*. Schereschewsky noted irregular forms of *Sp. pallida* in cultures, and was struck by the resemblance of some of these to *refringens*, and Sobernheim and Tomasoli also observed irregular forms. *Sp. pallida* is noted for the regularity of its curls and is indeed identified by this means; yet Schaudinn observed that almost straight forms might occur; Sobernheim and Tomasoli found specimens with straight portions toward the centre; and Fouquet recorded the discovery of straight forms in tertiary syphilis. Krzystalowicz and Siedlecki noted changes in *Sp. pallida* by which individuals contracted and thus became shorter and plumper. Eitner, who observed living forms indistinguishable from *Sp. pallida* noted that some of these straightened out when dead and were thus differentiated.

Krienitz studied the spirochætes present in the stomach and found that they underwent a change of form according to alterations in their environment. He

consequently held that morphology does not constitute a valid test of species.

These observations are sufficient to indicate that considerable doubt must be felt as to the constancy of form among the small spirochætes; and when it is remembered that the identification of such a form as *Sp. pallida* depends entirely upon its morphology, and that many forms have been described in conditions other than syphilis which can only be differentiated by experts from this spirochæte, the uncertainty that must continue to prevail as to relations of the small spirochætes to each other and to the morbid conditions with which they are associated, is undeniable.

HABITAT.

The only free-living spirochætes at present known are *Sp. plicatilis* and *Sp. daxensis* which are found in fresh water. *Sp. polyspira* is a saprophyte found in decaying vegetable matter (potato). The rest of the large spirochætes are saprophytes or commensals in the alimentary canal of various shell-fish. Of the smaller forms many are apparently saprophytes on the surface of ulcerative lesions; others are also saprophytic within the intestines of various animals. A few only are definitely pathogenic, being capable of multiplying within the living tissues of an animal host and there producing poisonous products.

CULTIVATION.

Spirochætes have never been satisfactorily cultivated; that is to say, they have never been isolated in artificial media and passed from culture to culture. Many authors have stated that they have succeeded in producing multiplication of spirochætes on laboratory media. Thus Duval and Todd kept *Sp. duttoni* alive

in a mixture of yolk of egg and decoction of mouse-flesh for forty days. They observed the formation of long chains under these conditions. Williams grew this organism in defibrinated blood, and Norris, Pappenheimer and Flournoy cultivated *Sp. obermeieri* in citrated blood. Lebailly states that *Sp. pallida* multiplied when kept on pieces of infected liver, and Volpino and Fontana found that it even grew into pieces of normal liver placed in contact with the infected tissues. Borrel and Burnet cultivated *Sp. gallinarum* in blood. Levaditi cultivated *Sp. pallida* and also *Sp. refringens* in collodion sacs placed inside the peritoneal cavities of animals, but these cultures were not pure, being contaminated with great numbers of bacterial organisms. Beer claims to have cultivated *Sp. pallida*, anaërobically like a bacterium on bouillon mixed with ascitic fluid.

Leuriaux and v. Geets state that they cultivated *Sp. pallida* in a mixture of broth with cerebrospinal fluid taken from syphilitic patients; after incubation in this medium the precipitate formed was sown on pigs' serum, and colonies developed. Other writers state that they have obtained multiplication of these spirochætes by incubating portions of infected organs; and Levaditi, as previously mentioned, "cultivated" *Sp. pallida* in collodion sacs within the peritoneal cavities of monkeys, but found the resulting "cultures" incapable of inducing infection.

Recently successful attempts to cultivate *Sp. pallida* are reported by Schereschewsky and by Mühlens. The medium used was horse-serum, which was first heated to 58° to 60° C.—a point at which it threatens to solidify —and then kept at 37° C. for three days. A piece of infected tissue (*e.g.*, a bit of a gland taken from a patient suffering from syphilis) is then inserted deeply into the medium and kept under anaërobic conditions for five

days or so, when multiplication of the spirochætes is found to have taken place. An odour of sulphureted hydrogen is evolved, which may be compared with the fœtid odour produced by the organisms of Vincent's angina. Schereschewsky found that forms resembling *Sp. refringens* first developed and afterward characteristic *pallidæ*. Inoculation of animals with the cultures was unsuccessful.

Levaditi and Stanesco also record successes in cultivating *Sp. gracilis* and *Sp. balanitidis* in a somewhat similar way. In the case of the latter organism they first inoculated tubes of horse-serum and then made sub-cultures in collodion sacs containing this serum, placed in the serum tubes. They noted that multiplication of the spirochætes only occurred when the media were liquefied by the action of proteolytic bacteria—an example of symbiosis which they compare with that needed for the cultivation of amœbæ.[1]

Most recently of all Bruckner and Galacesco have grown *Sp. pallida* on coagulated ascitic fluid and have successfully inoculated rabbits with sub-cultures made a month after the material had been sown on artificial medium.

MULTIPLICATION AND DEVELOPMENT.

Much controversy has centred round the question of the *mode of division* of spirochætes, large and small. Perrin described longitudinal division in *Sp. balbianii*, and Keysselitz in *Sp. anodontæ;* and I have seen what appeared to be this process taking place in the latter. Schwellengrebel believes that transverse division is the rule in *Sp. balbianii*, and Schellack finds it in *Sp. anodontæ* (Fig. 28) and in other forms. Among the smaller

[1] The "symbiotic" bacteria probably serve as food for the amœbæ, which need solid pabulum.

spirochætes longitudinal division is described in *Sp. duttoni* by Mayer and by Newstead; transverse by Duval and Todd, and by Koch. In *Sp. obermeieri*, (Fig. 29), both longitudinal and transverse division are found (Mackinnon; Nuttall, Fantham, and Porter). In *Sp. pallida*, longitudinal division is upheld by Beer and by

Fig. 28.—Spirochæta anodontæ; transverse division. (Schellack.)

Krzystalowicz and Siedlecki (Figs. 30, 31); transverse by Levaditi and Yamamouchi. In *Sp. vesperuginis*, Nicolle and Compte observed transverse division, Gonder on the other hand longitudinal (Fig. 32). In *Sp. gallinarum*, Borrel saw transverse division, Prowazek, longitudinal (Fig. 33); the latter method is described

by Gonder in *Sp. hartmanni*, the former by Martin in the spirochæte of the horse and by Wenyon in that of the mouse. Forked forms which are not infrequently

FIG. 29. FIG. 30. FIG. 31. FIG. 32.

FIG. 29.—Spirochæta recurrentis (Sp. obermeieri); transverse division. (Schellack.)

FIG. 30.—Longitudinal division of Spirochæta pallida. (Krzystalowicz and Siedlecki.)

FIG. 31.—Spirochæta pallida; multiple transverse fission. (Krzystalowicz and Siedlecki.)

FIG. 32.—Spirochæta vesperuginis; transverse division. (Gonder.)

FIG. 33. FIG. 34.

FIG. 33.—Spirochæta gallinarum, longitudinal division: *a*, First stage; *b*, second stage (transverse fission ?); *c*, daughter form after division. (Prowazek.)

FIG. 34.—Spirochæta obermeieri; filamentary forms in the bug. (Klodnitzky.)

observed are held to point to longitudinal division, forms with attenuated central strand, to transverse multiplication; but the latter formation has been held by

the supporters of longitudinal division to represent the final stage in this process, just before separation of the daughter individuals takes place (Fig. 33).

Gross believes that spirochætes become bent upon themselves before undergoing transverse division, the close apposition of the two limbs of the bent organism

FIG. 35.

FIG. 36.

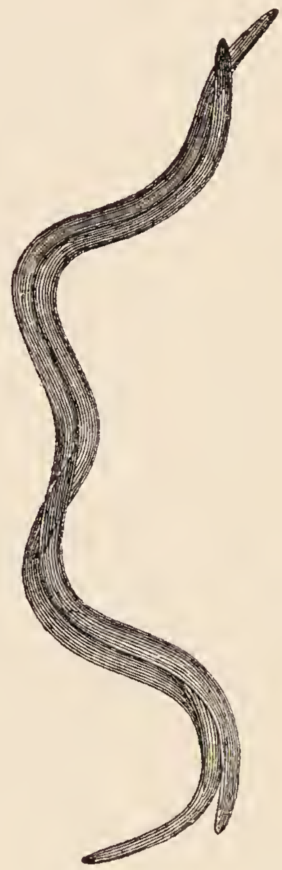


FIG. 37.

FIG. 35.—Spirochæta balbianii; supposed conjugation. (Perrin.)

FIG. 36.—Spirochæta vesperuginis; conjugation (?). ×2250. (Gonder.)

FIG. 37.—Spirochæta anodontæ; apparent conjugation, probably a stage in longitudinal fission.

then giving rise to an appearance of longitudinal fission. He terms the process division by "incurvation."

A development of the spirochætes into long *filaments* has been described by Duval and Todd in cultivations of *Sp. duttoni*, and by Klodnitsky as occurring in *Sp. obermeieri* within the body of the bug (Fig. 34). Koch also saw filamentary forms in ticks infected with *Sp. duttoni*.

The occurrence of *conjugation* in some form or other appears to be universal among protozoa, and if spirochætes are to be assigned to this class it will presumably be proved to take place among them also. At present the accounts given of such an occurrence are not conclusive (Figs. 35, 36). An instance which I observed of what appeared to be conjugation in *Sp. anodontæ* (Fig. 37) was probably only a stage in the process of longitudinal division. Krzystalowicz and Siedlecki have explained certain forked forms of *Sp. pallida*

FIG. 38.

FIG. 39.

FIG. 38.—Spirochæta pallida; formation of gametes. (Krzystalowicz and Siedlecki.)

FIG. 39.—Spirochæta pallida; conjungation. (Krzystalowicz and Siedlecki.)

which they observed as resulting from the conjugation of a small individual with a larger one (Figs. 38, 39); and Leuriaux and v. Geets describe microgametes and macrogametes. The four writers last mentioned agree in considering *Sp. pallida* to be in reality a species of trypanosome (*Trypanosoma luis*, Krz. and S.), the macrogamete being definitely trypanosomatoid and the microgamete spirochætal. Male, female and indifferent forms of *Sp. balbianii* are described by Perrin, but he failed to find any certain instance of conjugation (see Fig. 35).

Carter gives figures of what appears to be conjugation of a spirochæte which he found in human blood in

S. Arabia. Large globular swellings were formed in the bodies of the organisms and fusion seemed to take place between two individuals at these points (Fig. 40).

FIG. 40.

FIG. 41.

FIG. 40.—Arabian spirochæte; conjugation. (Carter.)
FIG. 41.—Spirochæta balbianii; encystment. (Perrin.)

The forms depicted are, however, so unlike those seen in any other spirochætes that it seems possible that Carter was dealing with a spirochætoid form of some other organism.

FIG. 42.

FIG. 43.

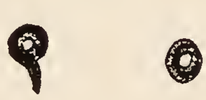


FIG. 44.

FIG. 45.

FIG. 42.—Spirochæta anodontæ, rolled up forms. ×500.
FIG. 43.—Spirochæta spiculifera, rolled up. (Schellack.)
FIG. 44.—Spirochæta pallida. Depression stage. ×2250. (Prowazek.)
FIG. 45.—Resting stage of Spirochæta buccalis. ×2250. (Prowazek.)

Encystment was believed by Perrin to take place in *Sp. balbianii*, and he gives illustrations of the movements of the organism in assuming this phase (Fig. 41). The

figures which he gives of the encysted stage are not very convincing. In the case of *Sp. anodontæ* I have found rolled up specimens after keeping the organisms for a day or two in the fluid which escapes from the mantle-cavity of the mussel (Fig. 42). These forms in some

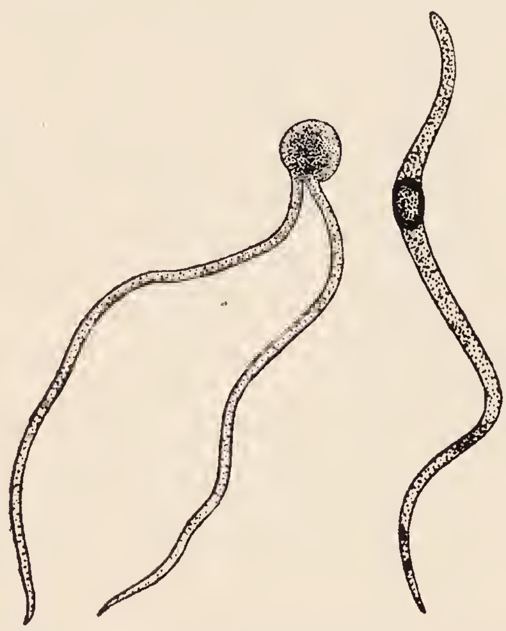


FIG. 46.—Spirochæta anodontæ; stages in formation of resting spore (?).

instances lay in or in close apposition to large epithelioid cells; but whether the spirochætes entered the cells at this stage, or the cell engulfed the spirochæte, or the apposition was merely due to some physical attraction, I cannot say (compare Figs. 69, 86, 87). The cells were

FIG. 46*a*.—Portion of Cladothrix putridogenes (Veszprémi) for comparison with Fig. 46.

necessarily somewhat degenerated after being kept so long in the fluid, and it seems unlikely that they would have been able to engulf the spirochætes.

Rolled up specimens of *Sp. duttoni* were seen by Mayer, and of *Sp. pallida* by Levaditi and others.

Oval forms were found by Leuriaux and v. Geets in *Sp. pallida;* they suggest that these develop subsequently into filaments. Ovoid bodies are also depicted by Krzystalowicz and Siedlecki as apparently constituting a stage in *Sp. pallida* (see Fig. 50), and somewhat similar bodies were found by Castellani along with *Sp. pertenuis.*

Sporulation.—In the case of *Sp. anodontæ* I have found some specimens showing about the middle of the body a rounded swelling or a darkly staining oval body

Fig. 47.

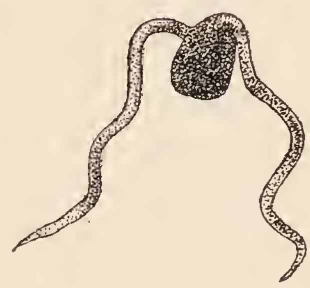


Fig. 48.

Fig. 47.—Spirochæta balbianii, with central swelling (so-called "male" form). (Perrin.)
Fig. 48.—Arabian spirochæte. (Carter.)

(Fig. 46) suggestive of a "resting spore." In figure 46*a* is depicted for comparison a portion of a filament of cladothrix showing a very similar swelling. These appearances may be compared with the illustrations given by Perrin of peculiar forms of *Sp. balbianii* (Fig. 47), and also Carter's figures of his Arabian spirochæte (Fig. 48).

Cox states that within the blood-corpuscles *Sp. obermeieri* breaks up into minute granules; and Leishman finds that *Sp. duttoni* gives rise to coccoid forms within the tick, no spirochætes being discoverable within ticks which are yet capable of conveying the infection. The so-called "moniliform" specimens of spirochætes (Fig. 27), are usually looked upon as degenerative forms but it is possible that the granules thus formed are capable of subsequent development, and are the equivalent

of bacterial spores. The terminal nodules of the smaller spirochætes are very suggestive of such a formation.

Associated with *Sp. anodontæ* are found chains of coccoid bodies, similar in length to the spirochætes. The arrangement of the staining material in the bodies of the latter, which may break up into regular portions

FIG. 49.—Development (?) of Spirochæta anodontæ: *a*, Spirochæte apparently undergoing segmentation; *b*, chain of cocci, possibly resulting from this process of fission.

of about equal size, suggests that they may give rise to the cocci by a process of multiple fission (Fig. 49).[1]

In this connexion it is interesting to note the occurrence of rod-shaped bodies, containing dots resembling nuclei, in association with different kinds of spirochætes. Thus they were found by Krzystalowic and Siedlecki along with *Sp. pallida* (Fig. 50) by Prowazek in connex-

[1] I hope to publish shortly further observations supporting the probability of this mode of development in *Sp. anodontæ*.

ion with *Sp. gallinarum* (Fig. 51) and with *Sp. lutræ* (see Fig. 71), by Mayer in the blood of a mouse infected with *Sp. duttoni* (Fig. 52), and by other observers elsewhere. Loewenthal found rod-shaped and sausage-

Fig. 50.

Fig. 51.

Fig. 50.—Abnormal forms associated with Spirochæta pallida. (Krzystalowicz and Siedlecki.)

Fig. 51.—Spirochæta gallinarum and rod forms. (Prowazek.)

shaped bodies along with spirochætes in ulcerated cancers.

Quéry believes that *Sp. pallida* is a stage in the development of a bacillus which he cultivated; from it he prepared a serum which Hallopeau found to have

Fig. 52.—Rod forms (*a*, *c*) and Spirochæta duttoni (*b*) in blood of mouse. (Mayer.)

some curative value in syphilis. Von Niessen also holds that the *Sp. pallida* is a developmental form of a special cocco-bacillus which he obtained in pure culture.

ASSOCIATION WITH OTHER ORGANISMS.

The association of spirochætes with other organisms is of considerable interest as either constituting an example of symbiosis or suggesting the possibility that

these organisms are a stage in the development of some other form.

The best-known example of such constant association is that of the spirochæte met with in Vincent's angina with the fusiform bacilli which are often regarded as the cause of this condition. The idea that the two forms may be stages in the life-history of one and the same organism is attractive and is supported by the experiments of Ruth Tunnicliffe who found that in cultures of *B. fusiformis* spirochætes made their

FIG. 53.—Spirochæta vincenti (*a*) with fusiform bacilli (*b*) and (?) filament of cladothrix (*c*).

appearance. This observation has not, however, been confirmed. Schmiedlechner found bacilli which divided longitudinally along with Vincent's spirochætes. Organisms resembling *B. fusiformis* were found by Mayer along with *Sp. duttoni* and by Krzystalowicz and Siedlecki with *Sp. pallida;* and Leishman, Harvey and Bousfield, and Launois and Loederich noted the coexistence of *Sp. pallida* with fusiform bacilli in syphilitic lesions. Wellmann found *Sp. pertenuis* along with fusiform bacilli and with *Sp. refringens.*

Other bacterial forms are also found associated with spirochætes, as by Dutton, Todd and Tobey in onychia, by Schereschewsky in syphilis (*B. pyocyaneus*), by

Cleland in castration-tumours in pigs, and by Cleland and by Bosanquet in ulcerative granuloma. Levaditi found it impossible to cultivate *Sp. pallida* and *Sp. refringens* in collodion sacs in the peritoneal cavities of animals without the simultaneous development of bacterial forms.

In gangrenous stomatitis the spirochætes are found most deeply situated in the tissues, the bacilli in the more superficial parts; while in ulcerative granuloma bacilli and spirochætes are found together deeply situated in the tissues, where it would seem unlikely that merely saprophytic germs would be able to establish themselves. The bacteria do not here resemble any known pathogenic variety.

The coexistence of *trypanosomes* with spirochætes was noted by Theiler in cattle, by Wenyon in mice, and by Petrie in birds (martins). Balfour found spirochætes in intestinal ulcers, in dogs and monkeys, produced by inoculation with trypanosomes (*T. dimorphon*); but as spirochætes or spirilla are common in the alimentary canals of animals, they may have been only accidentally present in these cases.[1] Gauducheau noted the appearance of spirillar bodies in a culture of amœbæ.[2]

The question as to the relationship between spirochætes and other organisms must at present be left undecided. In view of observations recorded in this and in the preceding section the possibility that spirochætes are only one stage in the development of a polymorphic organism must not be lost sight of as a probable explanation of this association, though it is at present far from proof.

[1]Balfour, *Jour. Trop. Med.*, 1906. [2]Gauducheau, *C. R. Soc. de Biol.*, 1908, LXIV, 493; Leishman, *Jour. R. Army Med. Corps* 1905, IV, 321; Petrie, *Jour. of Hygiene*, 1905, V, 195.

PATHOGENICITY.

Since it has been found impossible to produce pure cultures of any spirochætes, rigorous proof of their causal connection with disease, such as is demanded by the well-known "postulates" of Koch, cannot be furnished. Nevertheless, the pathogenicity of certain forms appears to be fairly established.

RELAPSING FEVER.

In the case of relapsing fever the spirochæte discovered by Obermeier and generally known by his name is universally admitted to be the exciting cause of the disease. It is present in all cases of the malady; it disappears during the remissions and reappears during the relapses. The blood of a patient reproduces a similar affection when injected into animals, and no other germ has been found present which might be looked upon as pathogenic.

The disease is characterised by a sudden onset, after an incubation-period of varying length (from a few hours up to fourteen days). The first access of fever lasts about a week and ends by crisis. A period of apyrexia ensues, to be followed by a second access of fever, and this sequence may be repeated a third and even more times. The spleen and liver are much enlarged, and the spirochæte may be found in the spleen during the remissions. Monkeys are the only lower animals directly susceptible to the disease.

In mankind infection may be conveyed by direct inoculation, as by a scratch at a necropsy; it may also occur by aërial conveyance in some instances, like the infection in scarlet fever, typhus, etc.[1] It has been supposed that the infection may be conveyed

[1] Ker, "Infectious Diseases" London, 1908, p 229.

from person to person by the bites of bugs, and it is probable that at least accidental infection may thus he produced, as Tictin found the spirochæte present in these insects and infected monkeys by injection of their macerated bodies; but it is not proved that an intermediate host is necessary or that any development of the spirochæte takes place in the bug. Manteufel and also Sergent and Foley believe that infection is conveyed by lice (*Pediculi capitis et corporis*).

The blood of a convalescent from relapsing fever contains substances capable of destroying the spirochætes (*spirillicidins*), and passive immunity is conferred by injection of such blood into another individual. Agglutinating substances are also formed. Metchnikoff holds that the spirochætes are mainly destroyed by phagocytosis; Rabinowitsch states that a hyperleucocytosis is produced in the blood of animals, and also a change in the bone marrow "like leukæmia."

Relapsing fever has been chiefly studied and described as it occurs in Europe especially in Russia. Closely allied forms of disease occur in other parts of the world, caused apparently by spirochætes which are morphologically almost indistinguishable from that of ordinary relapsing fever, but are probably different species.

AFRICAN TICK FEVER.

African tick fever is an affection met with in East Africa, and is conveyed by the bite of a species of tick, *Ornithodorus moubata*. The general features of the disease are similar to those of relapsing fever, but the relapses are often more numerous. The spirochæte present in this disease is said by Uhlenhuth and Haendel to be rather thicker than *Sp. obermeieri*, and by Schellack to move more vigorously. An attack confers

immunity against this parasite, but not against *Sp. obermeieri;* and agglutination tests also point to a specific difference between the two spirochætes. Mice and guinea-pigs can be inoculated with this organism, as can also rabbits, horses and monkeys. The pathological changes seen after death are the same as in infection with *Sp. obermeieri.* The spirochætes are taken up by the ticks from the blood of an infected person and are passed on in the egg to the young ticks of a second generation. Koch describes filamentary forms of the spirochæte in these animals. Levaditi and Manouélian believe that destruction of the spirochæte and cure of the disease are brought about by phagocytosis; some of the symptoms of the malady are attributed by them to thrombosis of blood-vessels caused by impaction of masses of spirochætes.

OTHER FORMS OF RECURRENT FEVERS.

Relapsing fever as seen in *America* is said to be caused by yet another spirochæte. Schellack gives the following description of the three allied organisms:

	Length	Breadth
African form .... (*Sp. duttoni*)	24–30μ (long forms)	0.45μ
American form ...... (*Sp. novyi*)	17–20μ (long forms)	0.31μ
Russian form........ (*Sp. obermeieri*)	19–20μ (long forms)	0.39μ

It is not impossible that other species may be found to exist in different parts of the world where forms of relapsing fever are encountered. Thus the

spirochæte of relapsing fever as seen in Bombay was found by Novy to differ from the American variety.

Spirochætes were also found by Carter in cases of fever occurring of S. Arabia. The illness consisted in headache, backache, prostration and enlargement of the spleen. Infection appeared to be conveyed by ticks (*Ornithodorus Sp. ?*). The relation of the disease to those described above is uncertain.

Darling believes that the spirochæte of relapsing fever in Panama is again a special variety, the immunity produced by an attack not being protective against the spirochætes of the other forms. The blood is infective in the intervals between the paroxysms. Apes and mice are susceptible to this parasite.

SYPHILIS.

The spirochæte discovered by Schaudinn in the lesions of syphilis and first named by him *Spirochæta pallida* is now generally accepted as the cause of this disease. Since it is not possible to obtain the organism in pure culture and to use such cultures for the inoculation of animals, rigorous proof of causal connection cannot be furnished. It is held, however, that as the spirochæte is constantly present in syphilis (100 per cent. of cases, Hoffmann and Prowazek) and is not found in any other conditions, it has as much claim to be regarded as the infective agent as has the *Bacillus lepræ* in leprosy. In view of its presence in the initial chancre, its subsequent passage to the neighbouring lymphatic glands, its presence in the blood in the secondary stage of the disease, and the occurrence only of degenerative forms in tertiary lesions (gummas) which are not supposed to be infective, the features of the disease may be satisfactorily explained as due to this organism. The discovery of immense quantities of spirochætes

in still-born fœtuses, the offspring of syphilitic mothers, is also urged as a strong argument in favour of the infective character of the organism. The serum of syphilitic subjects is said to agglutinate these spirochætes.

While we may admit that a strong *prima-facie* case can be made out for this view, it must be admitted that there are still some objections to be overcome before the *Spirochæta pallida* can be accepted as the proved cause of syphilis.

1. In the absence of cultures of the organism it can only be identified by its morphological characters. It is practically admitted that only those observers who have made a prolonged study of this spirochæte and of similar organisms can be sure of distinguishing it from other forms. This condition of affairs somewhat vitiates the statistical method of proof, dependent on the frequency of the finding of *Sp. pallida* in syphilitic lesions.

On the other hand, this difficulty of identification may be used as an argument by the up-holders of the spirochætal hypothesis against those who, like Kiolomenoglou and Cube, Krienitz and Scholtz, have found spirochætes which they identified as *pallidæ* in non-syphilitic conditions.

2. The question of the possible variability of form of *Sp. pallida* has already been discussed (page 30); if such variation be admitted, it becomes practically impossible to be sure that it is only in syphilitic lesions that this spirochæte is found, and its presence elsewhere would of course be fatal to its causal connection with syphilis.

3. Most observers have failed to find the spirochæte in all the cases of syphilis which they examined. The following figures may be quoted:

Author	Primary lesion			Secondary stage		
	Cases	Positive	Negative	Cases	Positive	Negative
Arning and Klein	112	108	4	179	169	10
Bandi and Simonelli	....	....	..	5	3	2
Bodin	16	10	6	17	10	7
Ferré	14	11	3	17	17	0
Kraus and Prantschoff	37	32	5	25	18	7
Metchnikoff	6	4	2	....	....	...
Mühlens	22	22	0	7	6	1
Shennan	10	5	5	....	....	...
Siebert	18	13	5	47	39	8

Bertarelli and Bovero examined forty-two cases and found the spirochæte present in twenty-six, absent in sixteen. They noted the scanty numbers of the organisms which could be found in the primary sore. Jaquet and Sevin were also struck by the difficulty of finding it in this position. Risso and Cipollina, and also Siebert, record the inconstancy of the organism in syphilitic lesions. There appears to be agreement that the spirochæte is not found in cases of "malignant syphilis" (see below). This is curious, since the more severe the disease, the greater the numbers of the pathogenic organisms which should theoretically be present. Even if these cases depend rather on a weakness of resistance on the part of the patient than on increased virulence of the spirochæte, there should still be plenty of the latter present.[1] The possibility that the condition is due to a mixed infection must, however, be borne in mind.

The relationship of the spirochætes to the actual lesions of the disease presents some points of peculiar-

[1] The title of a paper by Herxheimer (*Deut. med. Wochenschr.*, 1905, XXX, 1687) suggests that he found *Sp. pallida* in malignant syphilis, but the paper is not given in full.

ity. It would be natural to expect that in so well-marked a condition as the primary chancre, which appears to be produced by some intense irritant, there would be found large numbers of the causal organisms. As a matter of fact, spirochætes are almost invariably scanty in the tissues forming the chancre, and in many instances observers have failed altogether to find them. (Their presence on the surface of the ulcer is of comparatively little importance, in view of the ubiquity of similar organisms.) Others have failed to find the spirochæte in blood from a syphilitic case, which was definitely proved to be infective. The possibility of the existence of other stages of the parasite, *e.g.*, granules or coccoid bodies (p. 40), must, however, be borne in mind.

Again, spirochætes may be present in the organs of a syphilitic patient without apparently giving rise to any anatomical changes; as, for example, in an eye which appeared quite healthy (Bab). In considering the striking abundance of spirochætes in still-born syphilitic fœtuses it must be remembered that such bodies may be found full of other (bacterial) organisms, scattered throughout all the tissues; and also that Karlinski found spirochætes distributed throughout the tissues of a fœtal pig, having there presumably spread out from the intestine (see Fig. 57).

The presence of the spirochætes in gummas, which are not supposed to be infectious, is somewhat curious, for while this non-infectivity might be explained on the ground that the spirochætes here found are usually few and degenerate, yet if they are the cause of the disease at this stage, they must be of very notable virulence, since a gumma consists of a considerable mass of inflammatory tissue and may undergo rapid degeneration and softening, both presumably due to active microbial

action. So much irritation could hardly be attributed to the presence of only a few degenerate spirochætes.

4. The fact that the *Sp. pallida* is found in the lesions artificially produced by inoculation of lower animals with syphilitic material constitutes a two-edged argument, since the only animals which are definitely susceptible to syphilitic infection are the apes, while the spirochæte has been found living in dogs, cats, rabbits and guinea-pigs. It is necessary to assume that the pathogenic agent of syphilis can exist as a saprophyte in lower animals which are insusceptible to the disease—a condition which is at least unusual in the case of infective organisms. Otherwise it must be held to be saprophytic in both cases. In this connection the observations of Gaylord on the transmission of spirochætes in mice may be brought into comparison. This observer found spirochætes in certain cases of cancer in mice, and on inoculating other mice with emulsions of the tumours, he found that the spirochætes were also discoverable in the growths which resulted. The natural inference that the spirochætes were the cause of the cancer was, however, proved to be erroneous; and it appeared that the organisms were frequently present in mice and could be transmitted from one to another, but were harmless parasites of these animals. Of course, the parallel is not very close, since the *Sp. pallida* is not found in normal men or animals, but the possible transmission of spirochætes which are in no sense pathogenic must be noted.

5. The discovery of spirochætes, very like *Sp. pallida* in the closely allied diseases, yaws and ulcerative granuloma of the pudenda, would seem to constitute an additional argument in favour of the *Sp. pallida* being the true cause of syphilis. Herein also lies, however, a certain source of difficulty. It is generally said that the

spirochæte of yaws (*Sp. pertenuis*) is indistinguishable from *Sp. pallida.* So far as the silver-stained preparations made by Levaditi, for example, are concerned, this is true. (On the other hand, *Sp. pertenuis* as depicted by Castellani is different in appearance from *Sp. pallida*, not showing the same regular curls.) If then we have in yaws an organism exactly resembling *Sp. pallida*, the argument that this organism is only found in syphilis would seem to break down, since *Sp. pallida* is only recognised by its morphological characters. The identity of form is noted by Beurman and Gougerot, by Levaditi and Nattan-Larrier, by Schüffner and by Wellmann, and was admitted by Schaudinn himself.

6. Levaditi succeeded in cultivating *Sp. pallida* (*i.e.*, in causing some multiplication of the organisms and keeping them alive) in collodion sacs placed within the peritoneal cavities of monkeys. These cultures failed to produce infection when inoculated in other monkeys susceptible to syphilitic infection. The negative result is not perhaps in itself a very strong argument against the causality of the spirochæte, as the organism may easily have become attenuated by this procedure. Nevertheless it may be pointed out that Levaditi allows that a similar failure in the case of the spirochæte of balanitis is evidence against the causal connection of this organism with this disease, and the argument is as strong in one case as in the other. A positive result from inoculation of a culture has since been recorded by Bruckner and Galacesco.

7. The argument derived from the destructive and agglutinating action of the serum of syphilitic patients on the *Spirochæta pallida* is somewhat weakened by the fact that De Lisle and Jullien[1] found a coccus in

[1] *La Semaine Médicale*, 1900, page 279.

syphilitic patients which was agglutinated by their serum, but this organism is not now supposed to have any causal connection with the malady.

8. It is usually said that the result of antisyphilitic treatment is to cause the disappearance of the spirochætes; but Rille and Vocquerodt, and also Benda, found that no apparent effect was produced by treatment on the number of spirochætes present.

9. It is stated that whereas the syphilitic virus is active when infective material is mixed with glycerine, spirochætes are destroyed by contact with this reagent.

After a review of all the evidence at present available we can therefore only conclude that while there is a considerable probability that the *Spirochæta pallida* is the infective agent in syphilis, its position is not yet established with absolute scientific certainty.

The clinical phenomena of syphilis consist in (1) a primary lesion at the site of inoculation, consisting in an ulcer with a deeply indurated base; here the spirochæte is found on the surface of the ulcer and in the fluid that can be squeezed from its deeper layers, and it can also be found by the silver-staining method lying in the lymphatic spaces between the bundles of fibrous tissue. Enlargement of lymphatic glands takes place in the region towards which the lymph from the ulcer drains, and by puncture of these glands with a hypodermic syringe the organism can usually be obtained in the fluid withdrawn. (2). The secondary stage is marked by constitutional symptoms, such as fever and feeling of illness, along with eruptions on the skin and often with superficial ulceration of the throat (tonsils, fauces). In this stage the spirochætes may be found in the blood and in any vesicular lesions which may develop, or in the fluid obtained by blistering. (3). In a certain num-

ber of cases a tertiary stage is met with, consisting in the formation of localised inflammatory swellings, which may occur in any part of the body (the so-called gummas, from the "gummy" nature of the material formed by breaking down of the inflammatory cells). In these either no spirochætes are found, or only a few degenerate specimens. This stage is not infective.

Malignant syphilis is the name applied to certain forms of the disease in which ulcerative lesions appear at an early stage. Anæmia and cachexia develop to a severe degree, and the condition is not amenable to treatment with mercury. It "must be distinguished from severe syphilis which is characterised by dangerous localisation of the disease, or by association with other morbid conditions. The cause of malignant syphilis is doubtful; it has been attributed both to excessive virulence of the microbe and to secondary infection with other organisms. It would appear more probable that it is due to the implantation of the microbe on virgin soil—*i.e.*, on persons whose ancestors have been free from syphilis" (C. F. Marshall[1]).

Infection is usually contracted *in coitu* and is conveyed by the discharge from the primary sore. It can also be conveyed by means of drinking vessels or of a tobacco pipe recently used by an infected person in the secondary stage, in which there are infective lesions in the mouth and throat. Thus, either direct contact is necessary or very direct conveyance by an intermediary object. Some abrasion of the skin is probably required for the entrance of the infective organism, and surgeons are often infected through slight wounds in the fingers in the course of examining syphilitic patients. Infection does not seem to arise from contamination with the fluids of the dead body,

[1] "Syphilology and Venereal Disease," 1906, London, pp. 18–19.

as in making post-mortem examinations; hence it would seem that the organism quickly loses its virulence on the death of the host.

Congenital Syphilis.—Infection may be conveyed to the fœtus *in utero* from either parent. Whether the actual spermatozoon is infected cannot be demonstrated; certainly the causal organism can pass from the maternal blood, by way of the placenta, into the vessels of the fœtus. In this way congenital syphilis is produced. The syphilitic fœtus often dies before birth, and in such still-born fœtuses the *Spirochæta pallida* is found in practically all the organs of the body—most abundantly in the liver (see Fig. 88), and suprarenal capsules.[1]

It is generally supposed that resistance to the *Sp. pallida* is mainly effected by phagocytosis; but Zabolotny and Maskalowetz find that the serum of syphilitic subjects has an agglutinating and also a "lytic" power, so that the processes at work in producing recovery and immunity may be complex in character.[2]

Most attempts to pass the virus of syphilis through a porcelain filter have failed; but Jancke records one

[1] Some of the actual findings are: *In congenital syphilis*—in the eye and nasal mucus (Bab), in the blood and in the lungs (De Sousa and Pereira), in the bronchial epithelium and in miliary gummas (Benda), in the bullæ of pemphigus (Hoffmann, Leixer, Levaditi), in the testicles (Fouquet), in osteitis (Bertarelli), in the urine (Huebschmann), in the meconium (Simmonds), and in the placenta of the mother (Wallich and Levaditi). *In the acquired disease*, besides its localisation in the connective tissue of the primary sore, and in the lymphatic glands and in the blood in the secondary stage, it was found by Ehrmann in the nerve-sheaths, by Follet in the saliva. Veillon and Girard attribute the roseolar rash to the formation of embolic masses of the spirochætes in the blood-vessels. The organism may in rare instances be found in the cerebro-spinal fluid (Gaucher and Merle). It is also found in aortitis (Reuter, Schmorl). The spirochætes lie as a rule between the cells of the host; but they may be seen in polymorphonuclear leucocytes (Gierke) where they may be undergoing phagocytosis, and also inside the cells of the liver in congenital syphilis (Levaditi).

[2] The formation of a copula or amboceptor has already been noted. An account of the so-called Wassermann Reaction, based on this occurrence, does not fall within the scope of this little work.

positive result obtained with the material from a syphilitic fœtus.

YAWS.

The disease called yaws or pian (*Frambœsia tropica*) is in many ways like syphilis, going through a primary stage characterised by a local lesion at the point of inoculation, and a secondary stage of generalised phenomena, which take the form of multiple nodular inflammatory lesions or granulomas. It is generally supposed to be a distinct malady, but some writers still maintain that it is only a variety of syphilis. The *Sp. pertenuis*, first described by Castellani, is supposed to be the causal agent of the disease. It so closely resembles *Sp. pallida* (page 112) that some writers are constrained to argue from the diversity of the two affections to prove the distinct characters of the two parasites (*e.g.*, Wellmann).

Schüffner (who believes in the identity of yaws and syphilis) found the spirochæte present in 98 per cent. of all the cases he examined. Von dem Borne found it in seventy-three of seventy-six non-ulcerated cases: in ulcers it is found along with other kinds of spirochætes. Levaditi and Nattan-Larrier found the organisms on the surface of the primary lesion, in the glands, and in the spleen, but not in the blood. McIntosh states that the position in which this spirochæte is found in the skin is not the same as that in which *Sp. pallida* occurs; *Sp. pertenuis* being found in the superficial layers of the ulcer, in the fibrinous crust containing leucocytes and degenerated epithelium, whereas *Sp. pallida* occurs especially in the corium around the blood-vessels.

LYMPHADENOMA.

Proescher and White discovered spirochætes (*Sp. lymphatica*) present in a case of "lymphosarcoma."

With material containing it they inoculated a monkey (*Macacus rhesus*) and thus produced a local lesion at the point of inoculation, followed by the appearance of other nodules at a distance. They further inoculated a second monkey from the first, also with positive results. The spirochætes were found in all the lesions. These spirochætes so closely resembled *Sp. pallida* that they were at first mistaken (?) for this organism. It seems possible that in this case either a syphilitic adenitis was mistaken for lymphadenoma, or that a secondary infection with *Sp. pallida* occurred in a patient suffering from this disease.

ULCERATIVE GRANULOMA.

Spirochætes were first found in the disease called ulcerative granuloma of the pudenda by Wise, who stated that some forms resembled *Sp. pallida* and others *Sp. pertenuis*. They have since been found in this disease by MacLennan and by Cleland. They exist in the deep layers at the base of the ulcer, at some distance from the surface, and may be found in the blood-vessels. Bacteria are present along with the spirochætes. The pathogenicity of these organisms cannot be held to be proved as yet, but their position deep in the tissues is against a purely saprophytic character.[1]

VINCENT'S ANGINA.

Spirochætes are found along with fusiform bacilli in the disease called Vincent's angina, and also in some other conditions in which there is necrosis of tissue (*e.g.*,

[1] M. Carter states that he has found in a case of this disease, protozoon parasites in the form of minute rounded bodies within the cells. He believes that they are related to *Leishmania donovani* (*Lancet*, 1910, II, 1128).

in noma or gangrene of the cheek, in hospital gangrene, etc.). Uffenheimer thinks the spirochætes are the pathogenic agents in Vincent's angina, as, if the tonsils are removed in cases in which these organisms are present, further ulceration occurs. Ellermann also points out that the spirochætes are found more deeply situated in the tissues in cases of noma than are the bacteria, and that they seem to prepare the way for the latter. The relationship between the spirochætes and the fusiform bacilli has already been discussed (page 43). Veszprémi thinks that the spirochæte of Vincent's angina is identical with the common spirochæte of the mouth, *Sp. buccalis*.

OTHER DISEASES IN MAN.

Moritz found spirochætes in the bone-narrow and in the muscular coat of the intestine in a man who died with symptoms of intense anæmia, fever and diarrhœa, and in whom there were found after death an ulcer of the stomach and miliary nodules of new growth (endothelioma?) in the lungs. The primary seat of the tumour was not discovered. The spleen was enlarged. He thinks the spirochætes in this case were different from any previously described species, but their nature and relation to the disease must remain problematical.

Castellani found spirochætes present in certain cases of hæmorrhagic bronchitis (*Sp. bronchialis*). He believes that they may be the cause of the disease.

Waters also noted spirochætes in a series of cases of bronchitis, without hæmoptysis. Branch found organisms of this nature in a case of pulmonary tuberculosis, in which they were present in the expectoration along with tubercle bacilli; they may have been derived from the mouth.

Spirochætes are also supposed to be responsible for the diseases known as balanitis and ulcus tropicum (see pp. 107, 103).

DISEASES OF ANIMALS.

A number of diseases in the lower animals are associated with the presence of spirochætes. The best known is the so-called **spirillosis of fowls,** caused by the *Sp. gallinarum*. The disease is characterised by weakness, drowsiness and diarrhœa. It may prove fatal or recovery may ensue. The anatomical lesions met with consist in a fatty degeneration of the liver, with accumulation of uninucleated cells round the vessels, and hyaline necrosis in the spleen. The spirochætes are found in and around the vessels, in their walls, and between the cells of the liver. They do not usually become intracellular, but may be found in the mature ovules. Hereditary infection does not occur, the offspring of infected fowls being immune. The fertilised egg can, however, be artificially inoculated and then the chick is born infected. In such chicks the liver is first invaded and undergoes the change above described. Hæmorrhages also occur, and the blood is described as "myeloid" or "embryonic" or both together (containing granular myelocytes, vacuolated uninucleated cells, and nucleated red corpuscles with basophile granules).

The spirochætes are agglutinated by the serum of the infected birds. Levaditi considers that they are destroyed by phagocytosis, carried out especially by the macrophages of the liver and spleen. Neufeld and Prowazek deny this, and attribute recovery to the action of the serum, which is parasiticidal *in vitro* and also capable of protecting other birds. Gabritchewsky states that "lytic" bodies (bacteriolysins) appear in

the blood before phagocytosis occurs (in the goose), while Levaditi and Lange find exactly the reverse, the spirochætes disappearing before the formation of antibodies ("immobilisines") in the rabbit. Infection is conveyed by the bites of ticks (*Argas miniatus*, *A. reflexus*. *A. persicus*, *Ornithodorus moubata*, not by *Dermanyssus avium*). Atoxyl, a drug containing arsenic which is found useful in the treatment of trypanosomiasis, is also valuable in fowl spirochætosis, being both preventive and curative. Salvarsan ("606") is also very efficacious.

A disease of **cattle** characterised by fever and diarrhœa with enlargement of the spleen was described by Theiler. Crises and subsequent relapses may occur. Infection is conveyed by the bite of the tick *Rhipicephalus decoloratus*, and it would seem that some developmental stage may occur in this host, as direct injection of the blood of an infected animal into a healthy one does not result in infection.

Spirochætes were also found by Theiler in **sheep** and **horses** suffering from fever; he believes that these may have been the same organism as was found in the cattle. Baruchello and Pricolo found spirochætes in cases of infective pleuropneumonia in horses, the organisms being present in the spleen, in the pulmonary alveoli, in the pleural effusion, and in the blood. Martin also found spirochætes in a sick horse, which, however, recovered, and Stordy in a horse which died with symptoms of wasting and œdema; with these organisms the latter failed to infect a dog.

A disease due to a spirochæte (*Sp. vesperuginis*) was described in the **bat,** *Vesperugo kuhlii*, by Nicolle and Compte. Enlargement of the spleen was found in a

fatal case. Recovery by crisis and subsequent relapse were noted.

Dodd found spirochætes in certain lesions of the skin in a **pig,** infection being conveyed by contact. The organisms disappeared and reappeared during the course of the disease; they did not get into the blood of the animal, and inoculation was only possible in the skin. The lesions in some cases healed, yet the pig died. Anæmia and pneumonia were found *post mortem.*

Breinl and Kinghorn noted enlargement of the spleen in **mice** infected with *Sp. laverani.* This organism may be the same as *Sp. muris*, described by Wenyon, but the dimensions given by these writers are not identical, and Wenyon states that *Sp. muris* is a harmless parasite. He thinks it may be identical with *Spirillum minor* (*minus*) found by Carter in the **rat.**

SUMMARY OF THE PHENOMENA OF THE PATHOGENICITY OF SPIROCHÆTES.

Summarising the facts just recorded, we find that there is a group of spirochætes associated with affections in which the symptoms are *fever*, *a tendency to relapses*, and *enlargement of the spleen.* In this group we may place the spirochætes of the various forms of relapsing fever, the spirochætes of fowl-spirillosis, and perhaps those found in the bat. In these diseases the spirochætes give rise to a septicæmia—a generalised infection in which the organisms multiply in the blood-stream—without the formation of any local lesion. The causal connection of the spirochætes with these affections is clearly established.

On the other hand, we have in syphilis and yaws diseases in which there is a well-marked *primary local*

lesion, followed by a generalised infection. Relapses do not occur as a notable feature, and enlargement of the spleen is also inconstant. The causality of the spirochætes present is not yet absolutely proved.

In a third group, in which we may place Vincent's angina, ulcerative granuloma of the pudenda, and perhaps the disease recorded by Dodd in the pig, there is merely a *local affection* of the skin or of mucous membrane, conveyed in the last two cases by direct contact.

The three classes may be considered to constitute a descending scale of virulence on the part of the parasites, a local lesion being caused when the degree of virulence is slight, a local lesion with subsequent generalisation when it is rather greater, and an immediate septicæmia when it is most intense. Cases of congenital syphilis in which death occurs, but no anatomical lesions are discovered, have been attributed to a septicæmic action of the *Sp. pallida.*

METHODS OF STAINING.

The larger spirochætes may be stained by the ordinary methods used for protozoa—hæmatoxylin (Delafield's or Heidenhain's), Giemsa's stain, etc. They are not stained by carmine—thus resembling bacteria rather than protozoa.

For the smaller spirochætes, especially for *Sp. pallida*, a large number of methods have been recommended.

Giemsa's Stain.—This is made as follows: Azur II eosin 3 grm., azur II 0.8 grm., pure glycerine 250 grm., methyl alcohol 250 grm. One drop of this solution is added to 1 c.c. of distilled water. Stain for ten to fifteen minutes (see Plate, Fig. A).

Schereshewsky recommends diluting 13 to 15 drops of this solution with 10 c.c. of 0.5 per cent. solution of

glycerine. The mixture is warmed and, if no precipitate occurs, is poured on to the specimen and left for two or three minutes.

Schmorl uses Giemsa's stain for sections of tissue, following it by a watery solution of alum. He admits that this method is inferior to Levaditi's silver method.

Forest stains first with Ziehl's carbol-fuchsine in the cold, which dyes all spirochætes except *Sp. pallida*, and then stains with Giemsa's stain for twelve to sixteen hours in the cold and for one-half hour at 70° C.

Foix and Mallein note that the staining by Giemsa's solution may be hastened by the use of an electric current.

Other Methods.—Davidsohn recommends the use of cresyl violet for staining *Sp. pallida;* Oppenheimer and Sachs use carbolic gentian violet (concentrated alcoholic gentian-violet solution, 10 c.c., 5 per cent. solution of phenol, 100 c.c.). Proca and Vasilescu use Gino de Rossi's cilia-stain (dissolve 50 grm. pure phenol and 40 gr. tannin in 100 c.c. water and add to this 2.5 grm. basic fuchsine dissolved in 100 c.c. absolute alcohol. Stain in this for ten minutes, wash and dry. Then stain with a mixture containing concentrated alcoholic gentian violet 10 c.c., phenol 5 grm., distilled water 100 c.c.).

Reitmann advises that films should be first treated with 5 per cent. solution of phosphoric acid in water for five minutes and then stained with carbol-fuchsine, warmed.

Goldhorn uses a complicated preparation of polychrome methylene blue (methylene blue, lithium carbonate and eosin), and McNeal also uses methylene blue and eosin (crude methylene blue 20 parts, pure medicinal methylene blue 10 parts, eosin (yellowish) 20 parts, and pure methyl alcohol 100 parts. Stain on

cover-slip for forty-five to sixty seconds; wash in dilute solution of sodium carbonate—1 drop of 1 per cent. solution in 10 c.c. water).

Herxheimer and Hubner advise the use of bleu de Nil or bleu de Capri. Modifications of Jenner's or Romanowsky's stain are used by Simonelli and Bandi and by Von dem Borne. The former writers advise the method of May Grunwald, viz., dissolve 1 grm. of eosin (Grubler) in 1 litre distilled water, and 1 gr. of methylene blue (Meister, Lucius and Brüning) in a similar quantity of water; mix these solutions, allow to stand for two to seven days, and filter. Dissolve the sediment in pure methyl alcohol, and stain preparations with this solution for four to ten minutes.

Impregnation with Silver.—Bertarelli and Volpino first introduced this mode of staining spirochætes in tissue, but the modification of their method devised by Levaditi is usually preferred. His original method is as follows:

Fix fragments of tissue, about 1 mm. thick, in 10 per cent. formol solution; wash in water, and harden in 96 per cent. alcohol; then place in distilled water till they sink. Next place in the silver solution (silver nitrate 1.5 to 3 per cent.) at a temperature of 33° C. for three to five days. Wash in distilled water, and place in the reducing fluid (pyrogallic acid 2 to 4 per cent., formol 5 c.c., distilled water 100 c.c.) for twenty-four to forty-eight hours. Wash in distilled water, dehydrate, embed and cut (see Plate, Fig. B). A subsequent staining of the sections with Giemsa's stain or with toluydene blue may be carried out.

A subsequent modification of this method was devised by Levaditi and Manouélian, by which the first solution is made of 1 per cent. silver-nitrate solution with the addition of 10 per cent. pyridine; the tissues

are then washed in pyridine (10 per cent.); and reduction is effected by a fluid containing pyrogallic acid, 4 per cent., 10 per cent. acetone, and 15 per cent. pyridine in distilled water.

Ravaut and Ponselle use an albuminate of silver ("largine") in 2 per cent. solution, followed by 5 per cent. pyrogallic acid; the largine bath may be repeated a second time after the pyrogallic solution, the tissue being washed with distilled water between the two baths.

Yamamoto uses a 5 per cent. solution of silver nitrate, followed by a solution of 2 per cent. pyrogallic and 1 per cent. tannic acid. He prefers embedding in celloidin, and uses Loeffler's methylene blue as a counterstain.

INDIAN INK METHOD.

A special method of identifying these and other organisms on a field rendered opaque by means of indian ink has been devised by Burri. For this purpose ordinary indian ink is diluted with water (1 part in 6 or 1 in 10). The solution is sterilised and allowed to stand for two weeks, the supernatant fluid being then ready for use. A loopful of suspension of the organisms is mixed with a drop of the ink-solution, spread on a slide and allowed to dry. The spirochætes are then easily seen as colourless spirals on a dark background. Some writers prefer a stronger solution of the ink than that given above, *e.g.*, 1 in 2 of water.

EXAMINATION OF LIVING SPIROCHÆTES.

Dark Field Examination.—The identification of living spirochætes of the small varieties is most easily effected by examination on a "dark field" under the microscope, by means of one of the forms of par-

aboloidal reflector. The scrapings of tissue can be examined in physiological saline solution or in the fluid exuding from the sore, or a drop of blood can be mounted and sealed so as to exclude the air. The movements of the spirochætes render them easily visible. It is preferable, however, to add a drop of distilled water to the fluid containing the organisms, as this causes them to swell somewhat without losing their motility: they are thus more easily recognisable.

Intra-vitam Staining.—Mandelbaum stained living *Sp. pallidæ* in a hanging drop by adding a loopful of Lœffler's methylene-blue solution along with a loopful of decinormal soda-solution.

Meirowsky makes a paste of methyl-violet and salt-solution, and rubs it into the (previously cleaned) surface of a syphilitic chancre. In the serum which exudes there are found stained specimens of *Sp. pallida* and *Sp. refringens.* (Certain more deeply staining dots in the substance of the organisms he regards as nuclei.) Crystal violet is as efficacious as methyl violet. *Sp. dentium* may be stained by spreading a drop of fluid containing the organisms on a cover-slip, and placing this face downwards on a drop of concentrated watery solution of neutral red on a warm slide.

PSEUDO-SPIROCHÆTES.

A vigorous controversy was at one time waged as to the reality of the spirochætes seen in sections of tissue stained by the silver method, but there is now little doubt as to the genuine nature of these organisms. It may be conceded that the novice might be deceived by appearances presented by nerve fibres, by spirals of fibrin or elastic tissue, or by the edges of cells which take up the silver and present a crinkled appearance (Fig. 54); but those who have seen actual spirochætes

stained by this method could hardly confuse them with such objects or mistake the latter for them.

It is not often that a doubt arises as to the reality of

FIG. 54.

FIG. 55.

FIG. 56.

FIG. 54.—Pseudospirochætes; edges of cells. (Saling.)

FIG. 55.—Pseudospirochætes in a smear from the alimentary canal of Glossina palpalis. (From a specimen by Professor E. A. Minchin.)

FIG. 56.—Free flagellum of Trypanosoma lewisi.

spirochætes when other stains are used, but Professor E. A. Minchin has in his possession a film made from a "smear" of the contents of the alimentary canal of

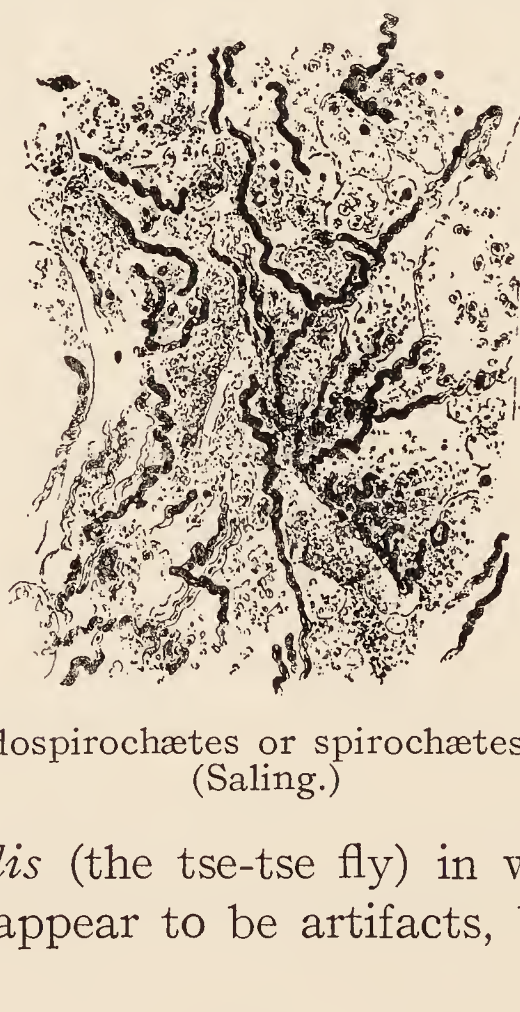


FIG. 57.—Pseudospirochætes or spirochætes in a pig fœtus. (Saling.)

Glossina palpalis (the tse-tse fly) in which are spiral objects which appear to be artifacts, but which have

almost exactly the appearance of true spirochætes (Fig. 55). The reason for regarding them as artifacts is that none were seen in other specimens of the same material and that their arrangement—often in crosses or radiating lines—is peculiar; but their resemblance is so exact that, if this view be correct, doubt may be thrown on many descriptions of spirochætes given by other writers.

Objects somewhat resembling spirochætes are the male gametes of some protozoa (coccidium, plasmodium), the flagella of trypanosomes which may occasionally be found free (Fig. 56), and attenuated forms of these last organisms and of Herpetomonas or Leishmania. The appearances found by Karlinski in a macerated pig-fœtus and figured by Saling (Fig. 57) may be either real or simulated spirochætes.

CLASSIFICATION.

It has already been pointed out that the position of the spirochætes in nature—whether they are to be regarded as protozoa or bacteria—is doubtful. Their relations as a group are almost equally undecided. The resemblance of the smaller to the larger spirochætes is not very close. Indeed, while each of these groups is fairly well marked, the relationship between the two is almost less striking than that of the smaller organisms to a group of bacterial organisms—the large spirilla. The *Sp. obermeieri* was originally classed with these, and it seems not impossible that this view is more correct than the recent classification of these little organisms with the spirochætes.

Certain peculiarities observed in the *Sp. pallida* led Vuillemin and afterward Schaudinn to separate it from the other small spirochætes and to put it in a distinct genus, called by the former *Spironema* and by the

latter (as this name was already occupied) *Treponema.* The characteristics of this genus were: The round shape of the body in section, as opposed to the flattened shape of the spirochætes; the fixity of the curls exhibited by the organism; the absence of an undulating membrane; and the presence of terminal filaments. *Sp. pertenuis*, from its close resemblance to *Sp. pallida*, was afterward placed in the same genus.

Examination of the qualities on which this distinction is founded does not tend to strengthen a belief in its validity. The round shape of the organism in section may be admitted, but the ribbon-like shape of the other small spirochætes is not very clearly established. The existence of an undulating membrane is doubtful even in the larger spirochætes, and still more problematical in the smaller group. Terminal filaments may be found in other species besides *Sp. pallida;* and finally the even curls presented by this spirochæte may at times be absent, as is admitted even by Schaudinn himself. Norris, Pappenheimer and Flournoy point out that the *Sp. obermeieri* exhibits all the characteristics assigned to a treponema.

Blanchard gives the following classification:

A. SPIROBACTERIA. Cohn, 1875 (Spirillaceæ, Migula, 1890). Curved bacteria; rigid; exhibiting transverse division.

1. *Spirosoma*, Migula, 1900. Curved rigid rod; no flagella. (*Sp. nasale*, Wibel, 1887. *Sp. linguale*, *id.*, 1888.)
2. *Vibrio*, Ehrenberg, 1878. (Microspira, Schrötter, 1886.) Rigid, flagellated rods, joined end to end, no spores. (*V. choleræ*, etc.)
3. *Spirobacillus*, Metchnikoff, 1889. Large spiral bacteria; lateral flagella; spores.

4. *Spirillum*, Ehr., 1833. Spiral, cylindrical organisms; blunt ends; no undulating membrane; spores; terminal flagella (many species).

B. TRYPANOSOMIDÆ. Doflein, 1901. Flagellated; flexible; undulating membrane; longitudinal division; not cultivable.

1. *Spirochæta*, Ehrenberg, 1833. (Spirochæte, Cohn, 1835.) Thin, spiral; flattened; undulating membrane; no flagella; filiform nucleus and chromatin granules (type, *Sp. plicatilis*).
2. *Treponema*, Schaudinn, 1905. (Spironema, Vuillemin, 1905). Characters as stated above. (*Trep. pallidum.*)
3. *Trypanosoma*, Gruby, 1844.

Lühe divides spirochætes into:

1. True spirochætes, including *Sp. plicatilis*, *balbianii*, *buccalis*, *dentium*, *vincenti*, *vaccinæ*, *refringens*, *pseudo-pallida*, and the spirochæte of dysentery.
2. Blood-spirochætes, including *Sp. gallinarum*, *anserina*, *obermeieri*, *duttoni*, *theileri*.
3. Treponema: *Tr. pallidum* and *pertenue*.

Gross who believes that spirochætes are closely allied to the bacteria, coins a new name for the whole genus, *Cristispira*, derived from the crest or ridge which he believes to run along one side of the body or around it (this formation having given rise to the statement that spirochætes possess an undulating membrane).

SECTION II.

SYSTEMATIC DESCRIPTION OF SPECIES.

The various spirochætes will be considered here in the following order:

LARGE FORMS.

Spirochæta plicatilis.
Sp. balbianii.
Sp. anodontæ and other spirochætes in shell-fish.
Sp. polyspira.

SMALL FORMS.

(Spirilla?)

Sp. obermeieri.
Sp. duttoni and other species in relapsing fever.
Sp. pitheci.
Sp. anserina.
Sp. gallinarum.
Sp. lagopodis. Sp. lovati.
Sp. vesperuginis.
Sp. lutræ.
Sp. theileri and allied forms.
Sp. suis.
Sp. laverani, Sp. muris, Sp. minor, Spirochætes in mouse-cancer.
Sp. gondii.
Sp. culicis, Sp. minei.
Sp. gadi, Sp. pelamydis, Sp. jonesii.
Sp. hartmanni.
Sp. bufonis.

Sp. buccalis, Sp. dentium, Sp. vincenti, Sp. gracilis.
Sp. schaudinni.
Spirochætes in gangrenous processes, hæmoptysis, etc.
Sp. microgyrata.
Sp. in small-pox and vaccine lymph.
Sp. refringens, Sp. balanitidis.
Sp. pseudo-pallida.
Sp. pallida.
Sp. pertenuis.
Sp. lymphatica and forms found in anæmia.
Sp. aboriginalis.
Sp. interrogans.
Various doubtful spirochætes and spirilla.

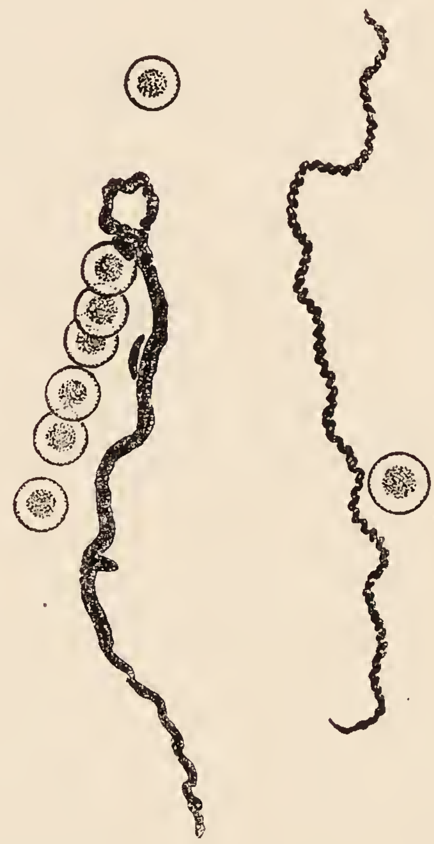
LARGE SPIROCHÆTES.

SPIROCHÆTA PLICATILIS.
(Ehrenberg, 1833.)

This, the first discovered species of spirochæte, is found in stagnant fresh water, especially along with algæ of putrefaction. It attains a length of 200μ, but its average length is 80μ and its breadth 0.5μ (Doflein).[1] In shape it shows a double series of spiral curves, a larger series producing a generally undulating appearance, and a smaller series imposed upon these (Figs. 1, 58). Its extremities are rounded, according to Bütschli; but Laptschinsky thinks that they are normally sharp and that blunt ends indicate that the organism has broken into pieces. It is flattened in form, like a ribbon. It possesses a periplastic sheath and contained entoplasm. Along the central axis is a refringent central rod, which stains darkly; granules of staining substance (nuclei) are arranged along this rod. This spirochæte, according to Bütschli, has no undulating mem-

[1] Illustrations of the organism make it appear much broader.

brane, Schaudinn who attributed to it such a membrane probably having mistaken for it the loose periplastic sheath. It exhibits screw-like movements, but also creeping and feeling (*kriechend*, *tastend*) movements, while the contraction of its sheath may lead to a thickening and thinning of the body at different places. Its mode of multiplication is not known, but it has been seen to break up transversely.



IG. 58.—Spirochæta plicatilis. (Schaudinn.)

The most recent description of this organism is due to Zuelzer, according to whom it is round in section, with a central axial thread of elastic substance and a simple spiral twist. A round refringent body is placed at the central point of each wave, and fine granules of volutin are also present. Sometimes a terminal granule is seen at one extremity, from which the axial filament starts. The length may reach 500μ and the breadth $\frac{3}{4}\mu$. The ends may be rounded or sharp. Transverse division occurs, either two or more frag-

ments being thus produced. A streaming of protoplasm may be seen. There is no undulating membrane or distinct periplast. The axial thread is important for classification. *Sp. plicatilis* may be found in the sea (Naples) as well as in fresh water. The organism is dissolved by trypsin.

As *Sp. plicatilis* is the "type species" and other spirochætes must justify their inclusion in the genus by their

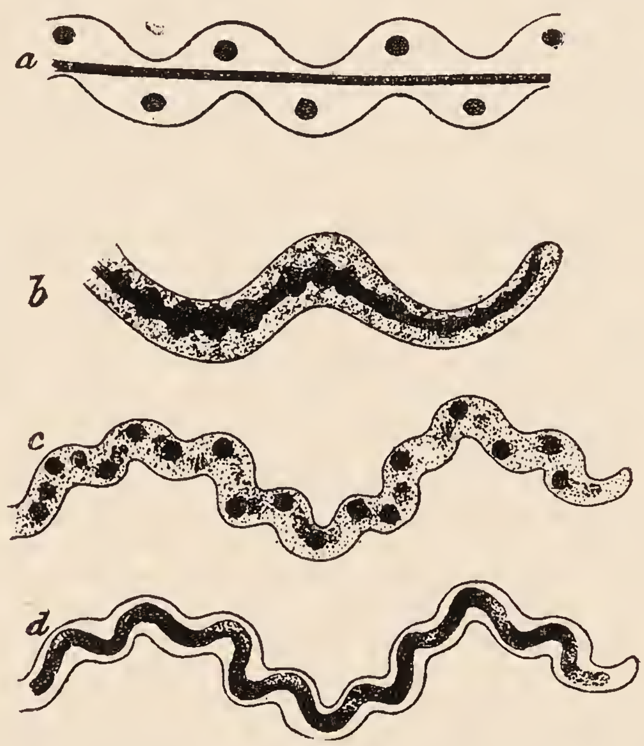


FIG. 59.—Diagrammatic representations of Spirochæta plicatilis; *a*, according to Zuelzer, straight central rod and dots in the curls; *b*, after Schaudinn, central rod follows the curves, dots superposed on it at random; *c*, after Doflein, double series of curls with corresponding dots and indications of central rod; *d*, imaginary figure showing central rod corresponding with dots in the diagram above.

resemblance to this, it is unfortunate that so great a diversity obtains between the descriptions given by various writers. Thus the organism is flat (Schaudinn, Doflein), round (Zuelzer); it has a double series of curls, smaller and larger (Doflein), a single series (Zuelzer); its axial filament follows the curves of the organism (Schaudinn); it runs in a straight line (Zuelzer). These differences are illustrated in the accompanying dia-

gram (Fig. 59). It is noteworthy that the nuclei (?) are distinct from the axial filament, whereas in *Sp. balbianii* the central staining rod is regarded as nuclear.

Very remarkable is the conflict of opinion between such observers as Bütschli and Schaudinn as to the possession of an undulating membrane by so large an organism as this. Its creeping and feeling movements are also noteworthy, as being unlike those seen in other spirochætes. The movements of swelling and thinning may be compared with the "euglenoid" movement seen by Plaut in *Sp. vincenti*.

It has been pointed out to me by Mr. T. P. Collings, to whom I am indebted for the preparation of the illustrations in this little work, and whose experience in drawing from the microscope is very extensive, that the series of dots seen in the illustrations of *Sp. plicatilis* is very suggestive of the existence of a spiral rod or filament seen in optical section in the tissues of the "cork-screw". Fig. 59, *d*, shows such a rod corresponding with the position of the dots in the diagram above it. The vast experience, however, of the authors who have described *Sp. plicatilis* makes it unlikely that they would have failed to recognise such a structure if it had been present.

SPIROCHÆTA DAXENSIS.

(Cantacuzène, 1910.)

Cantacuzène found in the water of the hot springs of Dax (52° to 56° C.) a spirochæte closely resembling *Sp. plicatilis*. It was from 30 to 100μ in length by $\frac{1}{2}$ to $2\frac{1}{2}\mu$ in breadth, flattened in form, and exhibited a double series of curls, smaller waves being superposed on the larger undulations. It possessed a longitudinal chromatin filament and was narrower toward the extremities, but these were not actually pointed. The

organism creeps about on the surface of the algæ present in the water. Cantacuzène distinguishes three kinds of individuals: 1, slender forms ($\frac{1}{2}\mu$ in breadth), the axial filament of which did not extend quite to the extremities; 2, shorter, thicker forms (1 to $1\frac{1}{2}\mu$ in breadth), exhibiting no axial filament and sometimes occurring in small masses; and 3. a few specimens showing a central swelling, some $2\frac{1}{2}\mu$ in thickness, in which all the chromatin was collected, though the rest of the organism stained darker than did the other forms. Sometimes two swellings were seen, one to the right and the other to the left of the central point.

SPIROCHÆTA BALBIANII.

(Trypanosoma balbianii, Certes, 1882.)

This large spirochæte was first described by Certes in 1882, though it had been previously seen by Moebius. It is found in the crystalline style of the oyster (*Ostrea angulata*, *O. edulis*). It attains a length of 100 to 120μ and a breadth of 5μ (extremes 26 to 100μ in length; 3 to 5μ in breadth, Perrin). Perrin describes an indifferent form, a female form, and male and female gametes. Taking the so-called *indifferent form* as typical, the general proportions being those described above, the organism consists of a periplastic sheath with fluid protoplasmic contents. The former is of somewhat firm consistency, retaining its form when the contents have escaped. It is furnished with an undulating membrane, running spirally round the body (Fig. 2). This has a dark peripheral fibre (border fibre, *randfibrille*) or thickening of the edge of the membrane. There is a dark nodule at each end of the periplast.

The nucleus consists of a spiral band (karyosome) formed of achromatic substance with bars of chromatin

at regular intervals; sometimes the bars may exist separately without the connecting band (Figs. 7, 60).

Multiplication takes place by longitudinal division, and may be very rapid in suitable conditions, giving rise to the appearance of very attenuated individuals, scarcely visible under the microscope. Division of the undulating membrane precedes that of the body of the organism. When fission is nearly complete, the daughter individuals remain attached by a strand of periplast for some time, appearing as a single individual of abnormal length with a thin portion in the middle. In the process of division the nucleus first condenses to form a straight rod, lying in the central axis of the animal. This rod breaks up into short segments, shaped first like bacilli and then as dumb-bells; these are next broken up into little spherical masses, which take up positions in two rows and finally in groups of four. Perrin believes that the total number of these is sixty-four. When fission has occurred and the new individuals are formed, the nucleus of each reforms a spiral band.

FIG. 60. Spirochæta balbianii. (Schellack.)

Encystment takes place by a series of movements by which the two ends of the spirochæte first travel alternately up and down its length, and then the whole organism rolls up into a coil (Fig. 41). The periplast then bursts, and the entoplasm escapes, taking a spherical form, but not secreting any covering membrane.

The *female* forms are described as larger and stouter, with a thicker periplast. They have a smaller undulating membrane, and encyst less readily. The nucleus goes through a somewhat different course of changes in

encysting. Masses of chromatin are formed, connected by thin threads. These divide into fragments, of which the greater number degenerate, but two remain and conjugate (autogamy), subsequently giving rise to the female gametes.

The *male* gamete is distinguished by the appearance of a rounded swelling of the entoplasm in the middle of the body, followed by longitudinal division of the organism, starting simultaneously at both ends. A reduction of chromosomes takes place in the central swelling, thirty-two remaining. Presumably the resulting individuals conjugate with female forms, but Perrin notes that he was able to observe only a few doubtful examples of cojugation (Fig. 35).

Fantham confirms Perrin's account of the general form of the nucleus. Further confirmation is needed of the different forms distinguished by Perrin, and of their true meaning, but we may compare Gonder's account of *Sp. pinnæ*. It is noteworthy that Perrin found that not all these spirochætes had undulating membranes, and also that he observed little difference in the movements of the forms with it and those without it. He does not accept the view of Laveran and Mesnil that the organism is furnished with a sheath, rather than with an undulating membrane, but agrees with them that it is a spirochæte, though he describes it as a trypanosome.

SPIROCHÆTA ANODONTÆ.

(Keysselitz, 1906.)

This large spirochæte was discovered by Keysselitz in the fresh-water mussel (*Anodonta cygnea*); it also occurs in *Anodonta mutabilis*. It exists in the crystalline style, and may also be found in the liver and in

the gastric epithelium. It attains a length of 130μ and a breath of 3 to 4μ. It is said by Keysselitz, to have a flattened form and either sharp or blunt extremities (Figs. 61, 62, 62*a*). Those with sharp ends show periplastic appendages (*periplastfortstaze*). The periplast is fibrillary in appearance, and there is a granule (*basalkorn*) at each end and an undulating membrane. The nuclear chromatin takes the form of balls, irregular masses and elongated bands; it may collect in a single strand toward the centre of the organism. Keysselitz

FIG. 61.—Spirochæta spiculifera. (Schellack.)

did not observe a spiral arrangement like that described by Perrin in *Sp. balbianii*, but admits that such may exist. Division is by longitudinal fission.

Schellack notes that the border fibre of the undulating membrane is of variable thickness, tapering toward the end, unlike that of trypanosomes, and believes that it is not a true membrane, but an artifact due to separation of periplast. I am inclined to agree with this view, as I find that the better the preparation is fixed the fewer organisms appear with this formation (see page 20). The nucleus, according to Schellack, consists of a series of bands, not of a spiral, and the whole

organism is built up of a series of chambers arranged longitudinally. Rolled up forms occur which may possibly represent cysts. He also believes that multiplication is by transverse, not by longitudinal division (Fig. 28), but I have seen a spirochæte apparently in process of longitudinal fission.

The blunt and pointed forms described by Keysselitz are held by Schellack to constitute different

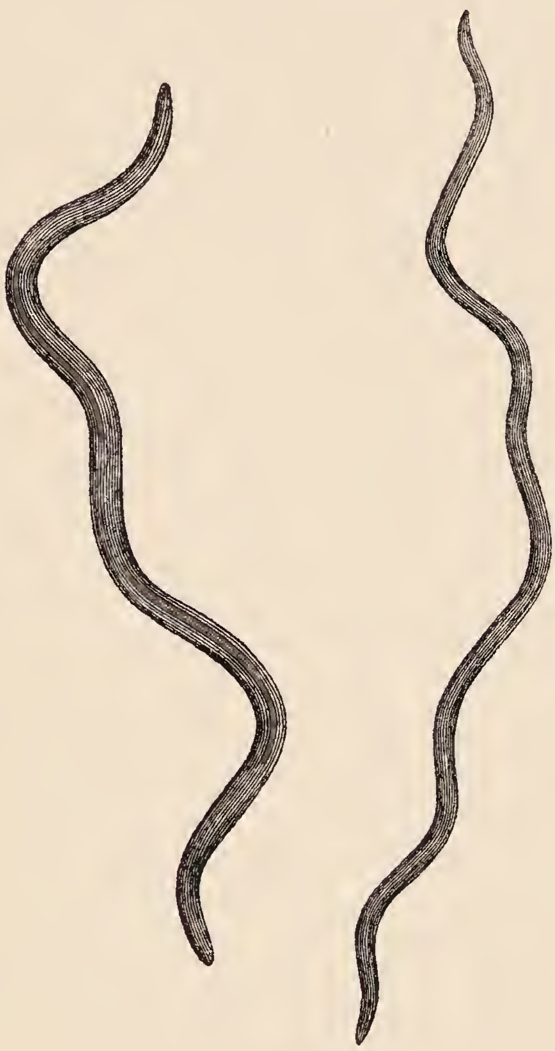


FIG. 62.

FIG. 62*a*.

FIG. 62.—Spirochæta anodontæ; specimen with blunt extremities; different sizes.

FIG. 62*a*.—Spirochæta anodontæ (Sp. spiculifera) forms with pointed ends; different sizes.

species; to the latter he gives the name *Sp. spiculifera* (Fig. 61). He describes a long form, from 39 to 50.5μ long and 0.9 to 1.2μ broad (average, 46μ by 1μ), and a sharp-pointed form, 28 to 36μ long and 0.7 to 1.1μ broad (av. 32μ by 0.9μ). In my own experience fourteen specimens with blunt ends gave an average length of exactly 100μ (extremes, 130 and 76μ), while of fifteen sharp-pointed specimens the average length was 55.6μ

(extremes, 96 and 34μ). The short variety tends to be rather thicker than the long. I find, however, spirochætes differing greatly in length and even more remarkably in thickness, which it would be difficult to sort out into two distinct species. I am inclined to regard the specimens with pointed ends as having recently divided. Schellack admits that it is curious that two forms should be found to coexist in each of three

FIG. 63.—Small spirochætes from the alimentary canal of Anodonta.

different molluscs (Anodonta, Tapes, and Ostrea), and there does not seem any good reason for making separate species of these slightly divergent forms at present. On the other hand, without stronger grounds we are not justified in describing them even as *sexual* forms of the same organism.

I have found in the alimentary canal of Anodonta minute spirochætes, 10 to 12μ in length (Fig. 63) which may be identical with *Sp. hartmanni* of Gonder or with *Sp. pusilla* of Schellack (see below).

SPIROCHÆTES OF OTHER MOLLUSCS.

Schellack has examined a large number of different shell-fish and found spirochætes in many of them, as shown in the following list. He believes that the average measurement of individuals taken just after division is a good criterion of species. He depicts many spirochætes with a line of separation in the middle of their length, pointing to transverse division. I have not myself been able to observe such specimens in the case of *Sp. anodontæ*, and even in the longest in-

LIST OF SPIROCHÆTES FOUND IN SHELL-FISH. (SCHELLACK.)

Name	Host	Length		Breadth		Ends
		Average	Extremes	Average	Extremes	
Sp. balbianii	Ostrea edulis	39	35–42	1.3	1.1–1.5	Rounded; no terminal appendages
Sp. ostreæ	Ostrea edulis	41.5	38–42.5	1.1	1.0–1.3	Sharp; no terminal appendages
Sp. chamæ	Chama gryphoides Ch. sinistrorsa	45.6	45–46.5	1.4	1.3–1.5	Rounded; no terminal appendages
Sp. anodontæ	Anodonta mutabilis	46	39–50.5	1.0	0.9–1.2	Rounded; no terminal appendages
Sp. spiculifera	Anodonta mutabilis	33	28–36.5	0.9	0.7–1.1	Rather pointed; terminal filaments
Sp. modiolæ	M. barbata	37.5	36–40	0.8	0.7–0.9	Rounded; no terminal appendages
Sp. pinnæ	P. nobilis	30.4	29–31	1.0	0.8–1.1	Rounded; no terminal appendages
Sp. limæ	L. inflata, L. hians	37	35–41	1.4	1.0–1.8	Rounded; no terminal appendages
Sp. cardii papillosi	Cardium papillosum	19.1	18.5–20	1.2	1.1–1.4	Rounded; no terminal appendages
Sp. tapetos	T. decussata	34.5	29–35	1.3	1.1–1.4	Rounded; occasional terminal appendages
??	T. decussata	33.4	29–36	1.4	1.3–1.7	? ?
Sp. acuminata [1]	Tapes læta	47	43.5–49.5	1.0	0.9–1.1	Pointed ends; no terminal appendages
Sp. saxicavæ	Sax. arctica	31	30–32	1.7	1.6–1.8	Rounded; no terminal appendages
Sp. gastrochænæ	G. dubia	29	constant	1.2	1.1–1.3	One end blunt, one sharp; no terminal appendages
Sp. pusilla	Anodonta, Unio, Lima, Tapes, etc.	13	12–14μ	....	0.3–0.4	Sharp-pointed

[1] This name had been previously applied by Castellani to a spirochæte found in yaws. See page 113.

dividuals which I have seen no sign of division was discoverable. I have, on the other hand, noted the existence of thin forms, which look as if they were formed by longitudinal fission, as was noted by Perrin in *Sp. balbianii.* Hence the above criterion does not seem adequate for the differentiation of species. Fantham describes *Sp. balbianii* as occurring in Tapes—which points to the probable identity of *Sp. tapetos* and *Sp. balbianii*, and throws further doubt on Schellack's classification. The following table embodies the latter's observations (see also Fig. 64):

SPIROCHÆTA PINNÆ.

(Gonder, 1908.)

This organism occurs in *Pinna squamosa* and *P. nobilis.* It varies from 10 to 60μ in length and in breadth from ½ to 3μ.[1] One end is blunter than the other, and here is situated a blepharoplast. An undulating membrane is present. The nucleus may take the form of a single rod or of irregular masses, which are sometimes arranged in groups of four. The rod-form is uncommon and is seen in very motile forms, which Gonder regards as possibly male elements, the irregular masses then characterising female or indifferent forms. A concentration of all the chromatin into one rounded mass was sometimes observed. Such individuals may constitute involution forms. Encysted forms also occur.

SPIROCHÆTA MACTRÆ.

(Prowazek, 1910.)

Found in the crystalline style of the mollusc, *Mactra sulcataria* (Deshayes), in Japan, this organism is from 45 to 70μ in length by 0.8 to 1μ in breadth. It has an undulating membrane.

[1] The dimensions of this organism given by Schellack are somewhat different (see table above).

FIG. 64.—Spirochætes from mussels. (Schellack.) *a*, Sp. spiculifera; *b*, Sp. cardii papillosi; *c*, Sp. saxicavæ; *d*, Sp. tapetos (× 2250); *e*, Sp. spiculifera; *f*, Sp. pusilla; *g*, Sp. pusilla; *h*, Sp. anodontæ; *i*, Sp. limæ; *k*, Sp. ostreæ; *l*, Sp. modiolæ. × 1000 (except *d*).

SPIROCHÆTA PECTINIS.

(Cristispira pectinis, Gross, 1910.)

Gross found spirochætes free in the alimentary canal of the mollusc, *Pecten jacobæus*. They were 72μ long, and 1.5μ thick, round in section, with blunt ends. They had a ridge or comb (*kamm*, *crista*) running along one side, but no terminal filaments. They seemed to consist of a series of chambers placed one behind the other, like a filament of some alga.

SPIROCHÆTA INTERROGATIONIS.

(Cristispira interrogationis, Gross, 1910.)

This organism was also found by Gross in *Pecten jacobæus*. It is 25μ in length by 0.5μ in breadth, and has pointed ends, bearing apparently to *Sp. pectinis* a relation similar to that of *Sp. spiculifera* to *Sp. anodontæ*.

SPIROCHÆTA POLYSPIRA.

(Wolff, 1907.)

This organism was found by Wolff in putrid potato and grows on ordinary media. He describes two forms: one with sixty to seventy curls, 70 to 80μ long by 1.5 to 1.6μ broad, which multiplies by longitudinal division, and a second 140μ long by 0.25 to 0.3μ, with sixty to eighty curls. Wolff is inclined to class this organism as a treponema. It is placed here on account of its length, but should perhaps rather be included with the small spirochætes.

SMALL SPIROCHÆTES OR SPIRILLA.

SPIROCHÆTA OBERMEIERI.

(Cohn, 1877.)

(Sp. recurrentis, Lebert, 1874.)

According to the recent descriptions of Novy and Knapp and other observers this spirochæte or spiril-

lum is from 7 to 19μ in length, and perhaps $\frac{1}{3}\mu$ in breadth (Fig. 65). Long forms may be met with, up to 100μ (120μ, Popovitch). It multiplies by transverse division (see Fig. 29), usually binary, but it is possible that multiple fission may at times occur. It usually stains homogeneously throughout, with the exception that the ends take the dye rather more faintly. Terminal appendages or flagella may be present. It has no undulating membrane or lateral flagella. Tangled masses of spirochætes may at times be seen, or two or more individuals twisted together. Granules are occasionally visible in the length of the organism, and moniliform degeneration occurs (see Fig. 27). Cox found spirochætes inside the blood-corpuscles, where they broke up into sections and finally into granules.

FIG. 65. Spirochæta recurrentis (Sp. obermeieri). (Schellack.)

Norris, Pappenheimer and Flournoy state that they cultivated these spirochætes in citrated blood. The spirochætes can be kept alive for as many as forty days outside the body (Novy and Knapp), but multiplication under such conditions is doubtful. Destructive (spirillicidal) and agglutinating bodies are formed in the blood of immunised animals, and passive immunity is conferred by injection of their serum. Apes, rats and mice can be infected with these organisms, but not rabbits, sheep, goats, fowls or pigeons (Fraenkel).

Fuchsine, gentian violet, Giemsa's stain and Levaditi's silver method may be used for staining *Sp. obermeieri*.

It is said that infection with relapsing fever may be carried by the air. The bites of insects have also been assigned as a means of conveyance of the disease. Klodnitsky describes a development of the spirochætes into long filaments within the bodies of bugs; but

Nuttall suggests that he mistook the spermatozoa of the animal for spirochætes (see Fig. 34).

Sp. obermeieri is destroyed by glycerine (Gabritchewsky). The action of a variety of different reagents upon these organisms was studied and tabulated by MacKinnon.

Sp. obermeieri in some form or other is capable of passing through a Berkfeld filter.

The spirochætes met with in cases of relapsing fever in America, in Bombay and in African tick fever are slightly different from the species met with in Russian relapsing fever, the typical form of the disease. *Sp. duttoni* of tick fever is described below. The spirochæte of American relapsing fever (*Sp. novyi*) is more delicate than that of the Russian disease and shows more regular and closely-set curls (Fig. 66). The spirillum of Bombay fever (*Sp. carteri*) is thinner, less regularly curled, and forms loops and "figures of 8" (Novy and Knapp). Animals inoculated with one species become immune to subsequent infection with this form, but not to infection with the other species.

FIG. 66.—Spirochæta novyi. (Schellack.)

SPIROCHÆTA DUTTONI.

(Novy and Knapp, 1906; Breinl, 1906.)

This organism was first discovered by Ross and Milne and found independently by Dutton and Todd. It occurs in the disease known as tick fever on the east coast of Africa. Infection is conveyed by the bite of the tick, *Ornithodorus moubata*, which exists in great numbers in the huts of the natives. The spirochæte is from 14 to 16μ long, with six to seven spiral turns;

long forms may be found up to 45μ in length. It has a dark central rod of staining material, and a faint surrounding periplast. It often exhibits dark or refringent granules in its body. Multiplication is probably by transverse division (Fig. 67), but forked forms are found. Rolled up specimens are seen within leucocytes, but these are considered by Levaditi and Manouélian to be degenerative, and not resting forms. The cilia described as present on these spirochætes by some writers are probably artifacts (Mayer). Koch states that *Sp. duttoni* shows no chromatin granules, but Mayer describes a differentiation of nuclear matter and protoplasm (see Fig. 24). The latter writer found in an infected mouse, forms resembling *B. fusiformis*, which may be stages in the development of the organism. These may be compared with those seen by Krzystalowicz and Siedlecki in cases of syphilis (possibly stages of *Sp. pallida*).

Fig. 67.—Spirochæta duttoni. (Schellack.)

L. A. and R. S. Williams state that they cultivated *Sp. duttoni* in defibrinated blood at room temperature, but it is doubtful whether true cultivation was effected. Duval and Todd devised a special medium for this purpose (see page 31) and succeeded in keeping the organisms alive for forty days. Levaditi "cultivated" these spirochætes in collodion sacs in the peritoneal cavities of rabbits along with the serum of monkeys (*Macacus cynomolgus*) and found that vibrio-forms developed.

A form of spirillum fever was observed by Hodges and Ross in Uganda, which is believed by Moffat to be different from the tick fever of the Zambesi region. The spirochætes found by Hodges and Ross were

apparently very large from 36 to 40μ in length by 4μ in breadth.

Allusion has already been made (page 37) to the spirochætes found by Carter in S. Arabia. No detailed description of the organism is given, but the writer describes longitudinal fission as taking place and also a process of conjugation. Figures 40 and 48 are copied from his original paper and show some of the forms observed; but the nature of their appearance is so peculiar that it would be premature to draw definite conclusions from the observations thus far made.

SPIROCHÆTA PITHECI.
(Thiroux and Dufougeré, 1910.)

Thiroux and Dufougeré found spirochætes in monkeys (*Cercopithecus patas*) suffering from an affection characterised by irregular attacks of fever—a condition resembling relapsing fever in man. The organisms were 15 to 20μ in length by 0.25μ in thickness, and presented five or six curls. Rolled up forms were also seen. The blood of the monkey was infective between the attacks of fever, and the disease was readily transmissible to mice, less easily to rats. In mice the incubation period was two to six days, and death usually occurred on the twenty-fifth or twenty-sixth day afterward. The spirochætes were present in one case in the cerebro-spinal fluid, and the monkey in this instance suffered from epileptiform attacks. The authors point out a resemblance to the position and action of the trypanosomes of sleeping sickness.

SPIROCHÆTA ANSERINA.
(Sacharoff, 1890.)

Sacharoff found these organisms in geese suffering from a fatal malady, characterised by fever, wasting

and diarrhoea. The bird sits apathetic and will not eat. The joints of the feet are so tender that the slightest handling of them causes cries of pain. The disease is a septicæmia, masses of spirochætes being found in the blood. According to Sacharoff geese and ducks are the only birds susceptible, the incubation period of the malady being two days. The blood of the infected bird is spirochæticidal, and immunity is produced in this way, not by phagocytosis (Gabritchewsky). Sacharoff, however, saw spirochætes inside leucocytes. Gabritchewsky injected a horse with the infected blood and obtained a preventive and curative serum.

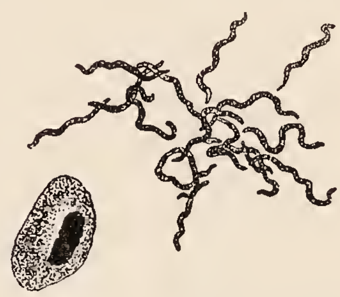


Fig. 68.—Spirochæta anserina with a blood-corpuscle for comparison. (Sacharoff.)

Cantacuzène holds that bacteriolysis and agglutination, in the case of these spirochætes, are phenomena which only occur *in vitro*, and states that destruction of the organisms takes place by phagocytosis in the spleen.

The spirochæte (Fig. 68) is from 10 to 30μ long and thinner than *Sp. obermeieri*. It multiplies by transverse division. A somewhat curious formation is depicted by Cantacuzène in organisms which are dividing, viz., the appearance of a faint rounded nodule in the centre at the point of division.

Borrel believes that *Sp. anserina* is the same as *Sp. gallinarum*, but this is not consistent with Sacharoff's statement that only ducks and geese are susceptible to the former.

SPIROCHÆTA GALLINARUM.
(Marchoux and Salimbeni, 1903.)

According to Prowazek these spirochætes have flattened bodies, terminal processes, and an undulating

membrane (?). He believes that they possess a central rod (*axenstrang*) which is elastic, and a contractile sheath. In length they vary from 4 to 20μ, and may show as few as two curls or a large number. Terminal granules are often seen, and the chromatin may gather to the centre of the organism, leaving the ends colourless. Division is longitudinal. Resting forms occur—some with terminal loops, others closely curled up, others again in the form of a "figure of eight" (see Figs. 26, 33, 51). They cling together in masses, but no true agglutination occurs, as the organisms free themselves again from the clumps. They may get inside the red blood-corpuscles (Fig. 69). Prowazek notes that the undulating membrane is best seen in macerated specimens. It is, therefore, probably an artifact. The cilia described by Zettnow as present on these organisms are also probably artifacts and due to plasmolysis. These spirochætes are killed by glycerine.

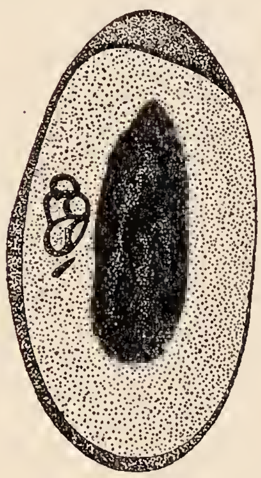


FIG. 69.—Spirochæta gallinarum in blood-corpuscle. (Prowazek.)

The spirochætes can be directly inoculated from fowl to fowl, and are also conveyed by the bites of ticks (*Argas miniatus*, *A. persicus*, *A. reflexus*, *Ornithodorus moubata*). Rabbits can be infected with this organism (Levaditi and Lange). Marchoux noted a loss of virulence in these spirochætes when they were passed through a series of fowls and thinks that the original virulence is regained by sojourn in the body of a tick.

The crisis of the disease is not due to the formation of antibodies, but may be due to phagocytosis. Atoxyl is both protective against the affection and curative when it already exists. Salvarsan (Ehrlich-Hata) is also very efficacious.

SPIROCHÆTA LAGOPODIS.
(Fantham, 1910.)

Fantham found this spirochæte in the blood of the grouse. It is from 10 to 18μ in length, and relatively broad (see Fig. 25). It is variable in form. Its ends taper to points, and it has an undulating membrane, spirally wound round its body. Within its substance may be seen a series of chromatin bars, and sometimes a helicoid core. It multiplies by both longitudinal and transverse division. It may be transmitted from one bird to another by the parasite, *Ixodes ricinus*.

SPIROCHÆTA LOVATI.
(Fantham, 1910.)

This organism is found in the cæcum of the grouse. It is from 16 to 32.5μ in length, and its ends are usually pointed, rarely rounded. It possesses an undulating membrane, and multiplies by both transverse and longitudinal fission.

SPIROCHÆTA VESPERUGINIS.
(Gonder, 1908.)

Nicolle and Compte found spirilla in a Tunisian bat (*Vesperugo kuhlii*). The organisms are from 12 to 18μ long, and $\frac{1}{4}\mu$ or less in breadth; they have pointed ends and no undulating membrane, and multiply by transverse division. Crisis and relapse occur, and there is enlargement of the spleen; some immunity is conferred by an attack.

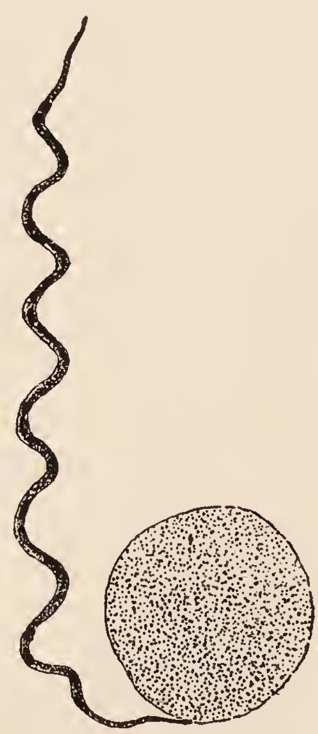


FIG. 70.—Spirochæta vesperuginis and blood-corpuscle. × 2250. (Gonder.)

Nicolle and Compte regard the organism as a bacterium, but Gonder claims it as a protozoon. The latter found individuals up to 30μ in length; and small forms, 3 to 5μ long, were

found in ticks (Fig. 70). He believes that this spirochæte divides longitudinally and has terminal appendages of periplast and an undulating membrane; but as he notes that this last only becomes visible on addition of glycerine, it is likely that the apparent membrane was an artifact. Gonder also describes dots of chromatin in the substance of the organism, which at other times are replaced by a chromidial network.

SPIROCHÆTA LUTRÆ.
(Prowazek, 1907.)

This organism was found by Prowazek in the blood of the otter. The only description which he gives of it is that it is a broad, band-shaped spirochæte, with blunt ends, and that large chromatin bodies (*Chromatinbrocken*) were visible in its protoplasm (Fig. 71).

FIG. 71.—Spirochæta lutræ and rod-shaped bodies. (Prowazek.)

Whether it had any relation to certain oval bodies ("like pébrine corpuscles") present in the blood along with it, he leaves undetermined.

SPIROCHÆTA THEILERI.
(Laveran, 1902.)

This organism was found by Theiler in cattle suffering from a disease characterised by fever, diarrhœa, and enlargement of the spleen. Disappearance and reappearance of the spirochætes may take place, as in the relapses of recurrent fever. The parasite is from

20 to 30μ long and $\frac{1}{4}$ to $\frac{1}{3}\mu$ in breadth; it appears in regular "cork-screws" and in irregular forms. It shows undulating movements, and may be free or attached to the blood-corpuscles. Infection may take place by the bite of the tick, *Rhipicephalus decoloratus*, and possibly a developmental stage may occur in this host, as direct inoculation did not succeed.

Theiler also found spirochætes in horses and sheep suffering from fever, and suggests that these may be the same organisms. He succeeded in inoculating a sheep with the bovine organism.

Martin found spirochætes in a sick horse in French Guiana; this form measured 12 to 15μ in length by $\frac{1}{4}\mu$ in breadth.

Baruchello and Pricolo found spirochætes in infective pleuropneumonia of horses; these organisms were apparently very small, measuring 3.2μ by 0.5μ. They occurred in the spleen, pulmonary alveoli, pleural effusion and blood. They sometimes exhibited terminal granules.

Heanley found spirochætes in buffaloes in China, and Martoglio and Carpano found organisms of this nature in sheep. The relationship between all these spirochætes is doubtful. Dodd believes that the spirochætes met with in the horse, ox, and sheep (called by Novy and Knapp *Sp. equi*, *Sp. bovis*, and *Sp. ovis*, respectively) are identical.

SPIROCHÆTES IN THE PIG.

(Spirochæta suis?)

Dodd found spirochætes in a disease affecting the skin of the pig, communicated by contact and inoculable in the skin of a healthy animal. The organisms (Fig. 72) were not found in the blood; they disappeared

and reappeared in the cutaneous lesions from time to time.

Cleland observed spirochætes in tumours occurring at the site of castration in pigs. The organisms were from 6 to 12μ in length and exhibited three or four irregular curls. Along with these organisms were large

FIG. 72.—Spirochæta suis. (After Dodd.)

bacillary forms, some of which were slightly undulating and suggestive of being modified forms of the spirochætes.

SPIROCHÆTA LAVERANI.

(Breinl and Kinghorn, 1906.)

These organisms, found in mice by Breinl and Kinghorn, measured from 1.8 to 3.75μ in length, and 0.1 to 0.2μ in breadth. They had pointed ends (one more so than the other) and one to four curls. They were transmissible by inoculation, with an incubation period of five days. Transmission by fleas and lice was not effected. The mice showed enlargement of the spleen.

SPIROCHÆTA MURIS.

(Wenyon, 1906.)

Found by Wenyon in mice, this organism (Fig. 73), is from 3 to 7μ long and 2μ broad. It has no undulat-

ing membrane, and multiplies by transverse division. The blood of the mouse is not infective, nor was transmission by fleas effected. It is apparently a harmless parasite.

Wenyon thinks it may be identical with the form found by Borrel in mouse-cancer and perhaps with the spirilla discovered in the rat by Carter.

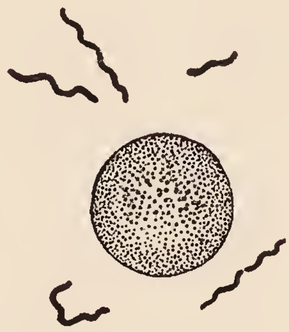


FIG. 73.—Spirochæta muris. (After Wenyon.)

SPIROCHÆTA MINOR.

(Spirillum minor, Carter, 1887.)

Carter found in the rat, *Mus decumanus*, spirilla (spirochætes ?) measuring 5 to 9μ in length, and possessing four to eight curls. He describes them as shorter and slenderer than *Sp. obermeieri*. They have pointed ends, and exhibit movements of rotation, propulsion, and lashing. They disappear and reappear in the blood of the rat, being replaced during their absence by granular particles and filaments.

SPIROCHÆTES IN MOUSE-CANCER.

Gaylord found spirochætes constantly present in mouse-cancers, both primary and secondary. They were studied by Calkins, who describes them as 2.5 to 7.8μ in length, 0.6μ broad, and possessing four to thirteen curls. The ends are blunt and rounded, and they have no undulating membrane or flagella.

Borrel had previously observed these organisms in 1905. They are not causally connected with the

tumours, but are passed from mouse to mouse in inoculating the growths. Calkins identified them with *Sp. microgyrata* (see below).

SPIROCHÆTA GONDII.
(Nicolle, 1907.)

Nicolle found spirochætes in the blood of the rodent, *Ctenodactylus gondi.* They measured 16 to 19μ in length by 0.3μ in breadth.

SPIROCHÆTA CULICIS.
(Jaffé, 1907.)

Jaffé found in the alimentary canal of the gnat spirochætes visible with a comparatively low magnification ($\times$ 220). They were flattened, band-shaped organisms, showing deeply stained granules in their protoplasm when treated with Giemsa's stain. Their movements were lashing, undulating, and "cork-screw-like." They were not furnished with an undulating membrane, and no examples of division were seen.

SPIROCHÆTA MINEI.
(Prowazek, 1910.)

These organisms are found in the stomachs of "worker" and "soldier" ants (*Termes lucifugus*, Rossi) in Japan. They are from 15 to 50μ in length, by 0.3 to 1μ in breadth. The extremities are pointed and chromatin granules are seen in the protoplasm. Division is longitudinal and rolled up forms and rosettes are found.

SPIROCHÆTA GADI.
(Neumann, 1909.)

This organism, found in the fish, *Gadus minutus*, resembles *Sp. gallinarum*, measuring 10 to 16μ in length,

with a wave-length of 3.5 to 4μ. It occurred both in the peripheral blood and in the internal organs, and was actively motile.

SPIROCHÆTA PELAMYDIS.

(Neumann, 1909.)

These spirochætes were found in the blood and organs of *Pelamys sarda*. They are shorter and thicker than *Sp. gadi*, measuring 9 to 10μ in length, occasionally as much as 18 to 20μ. The wave-length of the curls is 1 to 1.9μ. Movement is active.

SPIROCHÆTA JONESII.

(Dutton, Todd, and Tobey, 1906).

These spirochætes from the fish, *Clavias angolensis*, measured 18μ in length by 0.6μ in breadth. They appeared round at one end and pointed at the other, a feaure which suggests transverse division. They occurred both isolated and in clumps.

SPIROCHÆTA HARTMANNI.

(Gonder, 1908.)

This small spirochæte was found by Gonder in the intestine of *Pinna squamosa* and *P. nobilis*. It measures 6 to 14μ in length by 1μ in breadth. It is said by Gonder to multiply by longitudinal division, on the strength of certain forked forms observed. It has pointed ends and periplastic appendages.

SPIROCHÆTA BUFONIS.

(Dobell, 1908.)

This organism was found by Dobell in the rectum of the toad (*Bufo vulgaris*). It resembled *Sp. buccalis*, and measured 8 to 10μ by 1.5μ. It had an undulating membrane. The same writer describes another form

in the large intestine of the toad, which he compares with *Sp. pallida* and regards as a treponema, but the illustrations which he gives do not show the same regularity of curls.

SPIROCHÆTA BUCCALIS.
(Cohn, 1877.)

Several spiral organisms are found in the human mouth. The largest of these, *Sp. buccalis*, is from 12

FIG. 74.—Mouth spirochætes. (Spirochæta buccalis.)

FIG. 75.

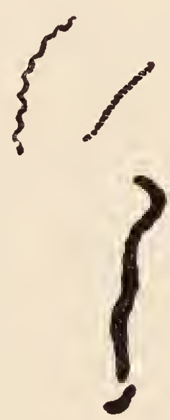


FIG. 76.

FIG. 75.—Spirochæta buccalis: *a*, *c*, Showing sheath; *b*, with terminal filament; *a*[1], Spirillum sputigenum. (Hoffmann and Prowazek.)

FIG. 76.—Spirochæta buccalis (below). Spirochæta dentium (above). $\times$ 2200. (Schaudinn.)

to 20μ in length and $\frac{1}{2}$ to 1μ broad (Figs. 74, 75, 76). It exhibits long undulations (wave-length 2μ, Mühlens and Hartmann) rather than short curls, and is active in

its movements. Goadby, however, describes spirochætes in the mouth, apparently *Sp. buccalis*, as having a sluggish oscillating movement. It has terminal periplastic appendages, and Hoffmann states that it has an undulating membrane; this is doubtful. Division is said to be longitudinal.

SPIROCHÆTA DENTIUM.
(Koch, 1877.)

This small spiral organism is from 4 to 10μ long, and exhibits well-marked regular curls (Figs. 76, 77, 78). Its breadth is about $\frac{2}{3}\mu$. It very closely resembles *Sp. pallida* in general appearance, but is said to be rather thicker, usually shorter, and to have rather less sharply-wound curls (Mühlens and Hartmann). It is uncertain

FIG. 77.

FIG. 78.

FIG. 77.—Spirochæta dentium. × 1500. (Hoffmann and Prowazek.)
FIG. 78.—Spirochæta dentium. Culture. (Mühlens.)

whether these organisms have any pathogenic influence; they are found chiefly in dirty mouths, in the tartar on teeth, and in the discharges of pyorrhœa; and Miller found tangles of them in a dental abscess, of which he thought that they were the cause.

These organisms may perhaps be the same as Loewenthal's *Sp. microgyrata* (see below).

An intermediate form, between *Sp. buccalis* and *Sp. dentium* is also described (*Sp. media* (Prowazek); (?) *Sp. denticola*, Arndt). Spirillar forms with lateral flagella (*Spirillum sputigenum*) also occur (Fig. 75).

SPIROCHÆTA VINCENTI.
(Blanchard, 1906.)

This spirochæte is met with in the affection known as Vincent's angina (a form of oral and pharyngeal ulceration described by Vincent) along with fusiform bacilli. The spirochætes are usually about 10μ long (extreme, 40μ, Mackie); they show rather sluggish movements (see Fig. 53). Plaut describes an euglenoid movement, or gradual swelling of the organism, passing like a wave along its length. These spirochætes are constantly associated with fusiform bacilli, of which some writers believe them to be a developmental form (see page 43). These or very similar associated organisms are found not only in the throat, but also in gangrenous processes in other parts of the body. The spirochætes have not been satisfactorily cultivated, but the bacilli may be grown on artificial media.

The resemblance of this organism to *Sp. buccalis* is very close, and some writers believe that they are identical (compare also *Sp. gracilis*).

SPIROCHÆTA GRACILIS.
(Veszprémi, 1907.)

This organism, found by Veszprémi in a case of abscess in connexion with the jaw, in a man, appears to be identical with *Sp. vincenti*. It occurred along with fusiform bacilli and cladothrix. Levaditi and Stanesco, however, obtained this organism (?) from a chancre and cultivated it by Schereschewsky's method. They state that it is a distinct species, almost indistinguishable from *Sp. pallida*: it exhibits, however, rather less regular spirals, is rather thicker, moves more actively, and is stained blue by Giemsa's method. The description is suggestive of *Sp. microgyrata* or *Sp. pseudo-pallida*.

SPIROCHÆTA SCHAUDINNI.
(Prowazek, 1907.)

Spirochætes and fusiform bacilli are found in the affection known as tropical ulcer of the leg (Fig. 79). Prowazek states that they are band-shaped organisms, possessing an undulating membrane, and that they multiply by longitudinal fission. In a specimen (from case of this disease?) sent to me by Capt. A. Whitmore, from Rangoon, the spirochætes were accompanied by a

FIG. 79.

FIG. 80.

FIG. 79.—Spirochæte of ulcus tropicum. Sexual forms. × 2250. (Prowazek.)

FIG. 80.—Spirochætes and cladothrix (?) in ulcus tropicum. (From a specimen by Capt. A. Whitmore.)

large number of threads resembling filaments of cladothrix (Fig. 80). The spirochætes were of about the same thickness as the threads, and stained almost exactly the same tint with fuchsine. It was difficult to resist the conclusion that they were forms of the same organism. (*Vide sub Sp. gracilis*).

Spirochætes and elongated rod-shaped bodies were found by Assmy in phagedænic ulcers.

SPIROCHÆTES IN GANGRENOUS PROCESSES.

Polland found spirochætes in cases of hospital gangrene; the organisms were usually from 15 to 20μ in length, but forms were met with up to 140μ. They

usually showed five or six blunt curves, and occurred along with fusiform bacilli.

Róna found spirochætes in noma, hospital gangrene, ulcus gangrenosum genitalium and pulmonary gangrene. They occurred along with fusiform bacilli. Those in noma and gangrene were of the same kind, but those seen in ulcus gangrenosum were smaller.

Possibly the latter were *Sp. microgyrata*, the former *Sp. vincenti*.

SPIROCHÆTA PSEUDOPALLIDA.

(Mulzer, 1905.)

This name was given by Mulzer to spirochætes closely resembling *Sp. pallida*, but not identical with it, found in sores on the genital organs. He thinks this is the same organism as was seen by Kiolomenoglou and Cube and by them described as *Sp. pallida*.

SPIROCHÆTA BRONCHIALIS.

(Castellani, 1907.)

Castellani found spirochætes in cases of Hæmorrhagic Bronchitis occurring in Ceylon. He noted the presence of four different kinds: (1) a thick form, with irregular curls, measuring 15 to 30μ; (2) a form like *Sp. refringens;* (3) a thin delicate form with tapering ends and small curls; and (4) another thin form with fewer curls. He speaks of an acute and a chronic bronchial spirochætosis.

Waters also found spirochætes associated with febrile bronchitis, and Branch noted two kinds in the sputum of a tuberculous patient (along with tubercle bacilli)—one resembling *Sp. pallida* and the other like *Sp. refringens*.

Bertarelli and Volpius found spirochætes resembling *Sp. buccalis* and others resembling *Sp. pallida*, but stain-

ing more readily, in the expectoration of a patient suffering from heart-disease.

SPIROCHÆTA MICROGYRATA.
(Loewenthal, 1906.)

Loewenthal found spirochætes in the discharges from the surfaces of ulcerated cancers. They stained more readily than *Sp. pallida*. Long individuals measured 5 to 11μ by $1\frac{1}{2}$ to 2μ; and smaller specimens $2\frac{1}{2}$ to 6μ by $\frac{1}{6}$ to $\frac{1}{4}\mu$. The small forms showed from four to twelve curls, with a wave-length of $\frac{1}{2}\mu$, or half that of *Sp. pallida*. Along with the spirochætes were rod-shaped and bent sausage-shaped bodies. Figure 81 shows one of these organisms as depicted by Schaudinn.

FIG. 81. Spirochæte of ulcerated cancer. (Schaudinn.)

Krienitz found *Sp. microgyrata* in cases of cancer of the stomach, and noted that the form of the organism changed with changing conditions, the alterations involving both length, thickness, and arrangement of curls. He therefore doubts the possibility of distinguishing spirochætes by their morphological characters alone.

Calkins identified the spirochætes found by Gaylord in mouse-cancer as a variety of *Sp. microgyrata* (see page 98).

Hoffmann found spirochætes in ulcerated cancers along with fusiform bacilli.

SPIROCHÆTES IN SMALL-POX.

Sakurane found spirochætes in the lesions of a patient suffering from small-pox, both in the contents of the pustules and in the tissues below these. They were long and slender, and showed numerous regular curls; forms with irregular curves were also seen.

SPIROCHÆTES IN VACCINE LYMPH.

Bonhoff reported the presence of spirochætes in vaccine lymph, but other writers believe them to have been artifacts.

SPIROCHÆTA REFRINGENS.
(Schaudinn, 1905.)

This spirochæte was found by Schaudinn in syphilitic lesions along with *Sp. pallida* (Fig. 82), but it may occur apart from the latter in simple lesions of the genital regions. It is broader than *Sp. pallida*, less

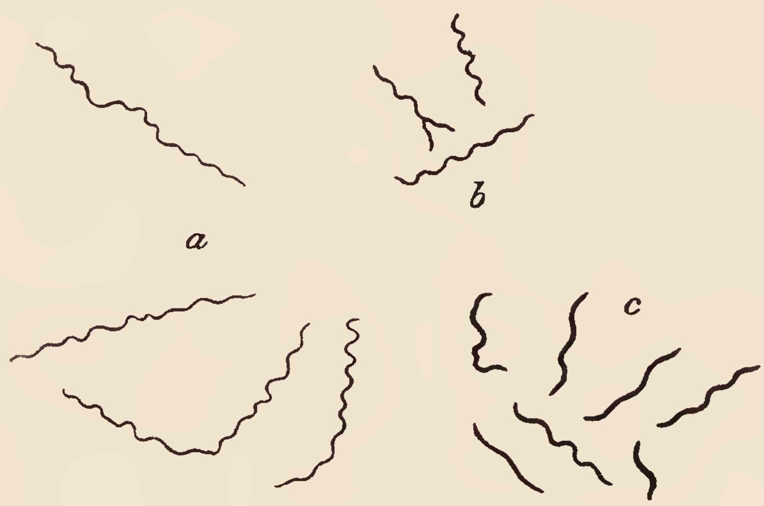


FIG. 82.

FIG. 83.

FIG. 82.—Spirochæta pallida and Sp. refringens. Smear from a syphilitic chancre; *a*, Sp. pallida; *b*, doubtful forms; *c*, Sp. refringens.
FIG. 83. Spirochæta refringens. (Schaudinn.)

sharply curled (Fig. 83), and stains more readily and deeply. Its length is from 8 to 12μ and its breadth about $\frac{1}{3}\mu$. It has terminal periplastic appendages, but no undulating membrane (Levaditi). Its body is said to be flattened (?), and not round like that of *Sp. pallida*.

Some writers believe that *Sp. refringens* is the same as *Sp. balanitidis* (Rille, Kraus). It may be the same organism as was described by Donné in 1837 as *Vibrio lineola* in syphilitic lesions. Eitner believes that more

than one species of spirochæte is included under the name of *Sp. refringens*. Richards and Hunt distinguished three forms of *Sp. refringens*, somewhat differing in appearance, and also believed that this organism might be a stage in the development of *Sp. pallida* (see page 30).

Sp. refringens was cultivated by Levaditi in human blood in collodion sacs placed in the peritoneal cavities of rabbits. The spirochætes developed, showing short vibrio-like forms along with long spirochætes; different species of bacteria also developed in symbiosis. Inoculation of these cultures failed to induce balanoposthitis, and Levaditi does not think the spirochætes are the cause of this disease.

Sp. refringens was found by Baermann in enlarged glands in a monkey infected with syphilis, along with *Sp. pallida*; it may therefore become a blood parasite, and not be confined to superficial lesions.

FIG. 84.—Spirochæta balanitidis. *a*, Terminal filaments and close curls; *b*, dividing; *c*, supposed undulating membrane. (Hoffmann and Prowazek.)

SPIROCHÆTA BALANITIDIS.

(Hoffmann and Prowazek, 1906.)

The causal (?) organism of balanitis is believed by Hoffmann and Prowazek to be distinct from *Sp. refringens*; they describe it as a band-shaped spirochæte (Fig. 84) with six to ten curls, $\frac{1}{2}$ to $\frac{3}{4}\mu$ broad, and furnished with periplastic appendages and an undulating membrane (the illustration showing the later formation is not convincing). It exhibits wave-like and rotatory movements. It is stained red by Giemsa's reagent. Scherber states that these spirochætes can be grown anaërobically on media containing serum;

he also finds that they make their way into the blood-vessels and are not confined to the surface of the ulcerative lesions, as is usually supposed.

SPIROCHÆTA PALLIDA.
(Schaudinn, 1905.)

(Spironema pallida, Vuillemin, 1905; Treponema pallidum, Schaudinn, 1905; Spiroschaudinnia pallida, Sambon, 1907.)

This organism was discovered by Schaudinn in March, 1905, and announced in a communication by Schaudinn and Hoffmann. It is usually from 4 to 14μ long, and exhibits six to fourteen curls; but longer forms are encountered, up to 40μ or more. Its breadth is almost unmeasurable, and may be $\frac{1}{4}\mu$ or less.[1] It has pointed

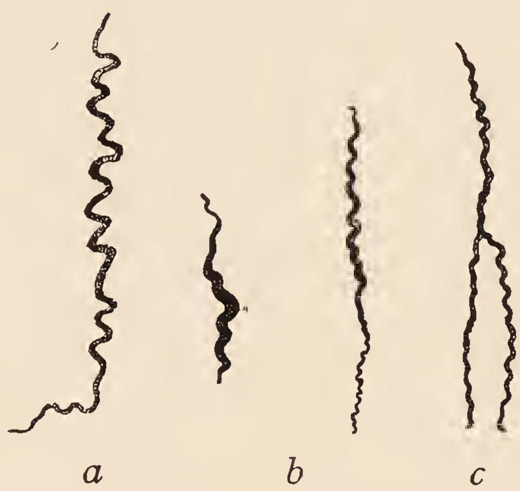


FIG. 85.—Spirochæta pallida: *a*, After Schaudinn; *b*, *c*, after Krzystatowicz and Siedlecki.

ends and terminal appendages of periplast, but no undulating membrane (Fig. 85). Its body is round in section, and the curls are very constant and regular. It differs from most other spirochætes in staining a reddish colour with Giemsa's stain, instead of blue. Its movements are rotatory and undulating, occasionally lashing, the curls remaining constant during these motions. In certain conditions, however, the curves

[1] They appear considerably thicker when stained by Levaditi's silver method than with ordinary stains.

are obliterated, and straight and other atypical forms are seen (Schaudinn). Such irregular forms are common in "cultures" (Levaditi and MacIntosh, Schereschewsky). They may closely resemble *Sp. refringens.* In movement the long slender organisms may become short and plump (Krzystalowicz and Siedlecki).

FIG. 86.—Spirochæta pallida in blood-corpuscle. (Gonder.)

Forked forms of *Sp. pallida* are found and also specimens showing apparently two individuals twisted together; from these appearances a longitudinal mode of division has been inferred, but Levaditi states that division is transverse. Refringent granules may be seen in the length of the organism, and also terminally placed.

The spirochæte is easily found by mixing a loopful of secretion from a syphilitic sore with a drop of distilled water on a slide, dropping on a cover-slip, and observing

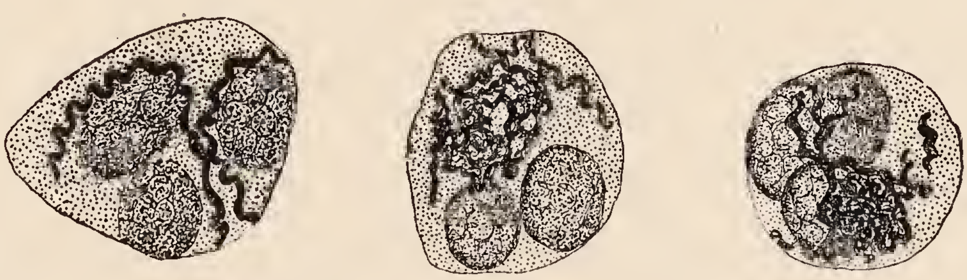


FIG. 87. Spirochæta pallida in leucocytes. (Gierke.)

in the "dark field" (see p. 66). Smears from such a sore may be fixed in osmic vapour and stained with Giemsa's reagent, or fixed and stained simultaneously with this fluid (see plate, Fig. A).

In the body the spirochætes lie chiefly between the cells in the lymphatic spaces, in lymphatic vessels and glands, and in the blood. Occasionally they become intracellular (Figs. 86, 87). The appearance of the organisms in the liver of a syphilitic fœtus, when stained

by Levaditi's method, is shown in figure 88 (see also plate, Fig. B).

A sexual cycle has been described by Krzystalowicz and Siedlecki. According to these writers, certain spirochætes become thicker and less sharply curved, and constitute the female gametes. Other individuals are found with many nuclei, and these by fragmentation produce a large number of minute spirillar forms, which are the male gametes (Fig. 38). One of these conjugates with a female gamete, and the individual

FIG. 88.—Spirochæta pallida in liver of syphilitic fœtus stained by Levaditi's silver method.

thus formed probably enters on a resting stage, which has not been definitely identified. These writers consider the *Spirochæta pallida* to be a trypanosome, and call it *Trypanosoma luis*. The same view is taken by Leuriaux and v. Geets, who describe oval bodies which elongate and give rise to fine filaments; these latter are the microgametes (the spirochætal form), while the female gametes are represented by a tryponosome-like stage. They suggest that the oval forms found by them are identical with Siegel's *Cytoryctes luis*.

The observations of Krzystalowicz and Siedlecki

have not been confirmed. Apart from the sexual cycle which they describe, it is clear that if the variations in the form of *Sp. pallida* described by them really occur, the foundations of the whole theory of its causal connection with syphilis are shaken, resting as they do upon the morphological constancy of this organism, with consequent facility of identification, and its occurrence only in syphilitic lesions. The observations of these writers would point to the existence of forms indistinguishable from *Sp. refringens* and possibly from other organisms.

Peculiar bodies were found by Krzystalowicz and Siedlecki in the fluid obtained by blistering with the actual cautery over a syphilitic papule; these were much larger than the ordinary spirochætes and showed irregular undulations and often an enlargement at one end. They were irregular in shape and seemingly amœboid. Their relationship to *Sp. pallida* was uncertain. They seem to be the same bodies as were found by Horand, who noted that they were twenty-seven or twenty-eight times the diameter of a red blood-corpuscle and had a head, neck and tail. Babes and Panéa also found in a case of congenital syphilis bodies resembling spermatozoa, having an elongated head, 1 to 2μ long, and a wavy tail or two tails. They were inclined to doubt whether these were parasites.

Forms somewhat resembling *B. fusiformis* were noted by Krzystalowicz and Siedlecki, but they do not seem to have regarded them as bacterial.

Many attempts have been made to cultivate *Sp. pallida*. Recently success has been obtained by Schereschewsky and by Mühlens; and Bruchner and Galacesco state that they inoculated rabbits with cultures and produced syphilis (see p. 33).

SPIROCHÆTA PERTENUIS.

(Castellani, 1905.)
(Spirochæta pallidula, Castellani, 1905.)

Castellani found spirochætes present in eleven out of fourteen cases of the disease called yaws, frambœsia, or pian; and the discovery was confirmed by other observers (Borne, Schüffner, etc.). The organisms are very slender, and vary in length from a few microns up to 18 or 20μ. Their extremities are pointed as a rule, though some individuals appear to have blunt ends. Their curls are numerous and symmetrical, but sometimes part of the organism may appear straight (Fig. 89). Nodules may be visible at their extremities, and

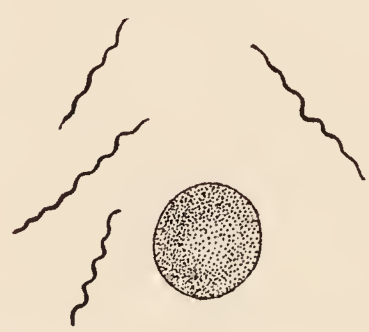


FIG. 89.—Spirochæta pertenuis. (Castellani.)

also in their length (Fig. 21). Ovoid bodies were also found by Castellani (5 to 8μ by 4 to 6μ) and were thought by him to be possibly a stage in the development of the spirochætes; they were very rare.

Monkeys were inoculated with the spirochætes, and the organisms were found in the resulting lesions. Monkeys thus inoculated showed no immunity to syphilis; this is held to prove the two diseases to be different.

These spirochætes are almost exactly like *Sp. pallida*, especially when stained with silver by Levaditi's method; but McIntosh states that the positions they respectively occupy in the lesions are different (see page

57). In the illustrations given by Castellani *Sp. pertenuis* does not appear so regularly curled as is *Sp. pallida*.

SPIROCHÆTA OBTUSA.
SPIROCHÆTA ACUMINATA.
(Castellani, 1905.)

Along with *Sp. pertenuis* Castellani found other forms of spirochætes which he called respectively *Sp. obtusa*—a thin delicate organism with blunt extremities—and *Sp. acuminata*, also very slender, but with tapering ends. Wellman could not identify these varieties in the lesions of yaws. Their nature and their relationships to *Sp. pertenuis* are doubtful.

SPIROCHÆTA LYMPHATICA.
(Proescher, 1909.)

Proescher and White found spirochætes in two cases of lymphadenoma; the organisms were present in enormous numbers in the glands, being demonstrated by Levaditi's silver method. A rhesus monkey was inoculated with material from the glands, and developed first a local nodule at the site of inoculation, and subsequently further secondary nodules in distant parts of the body. Spirochætes were found in all the lesions. A second monkey was successfully inoculated from the first. The spirochætes resembled *Sp. pallida* so closely that Proescher at first believed the case to be syphilitic.[1]

We may here mention a spirochæte found by Moritz in a case of severe anæmia and cancerous lymphangitis, the organisms being present in the bone-marrow and in the wall of the gut. With the spirochætes in the latter position were bacterial forms. The spirochætes were

[1] It is possible that there was actually a syphilitic infection in this case.

8

from 2 to 6μ in length, and had from three to ten curls. They differed from *Sp. obermeieri* in their appearance, being plumper and more compact (*gedrungen*); they had also thicker ends.

SPIROCHÆTA ABORIGINALIS.

(Cleland, 1909.)

Spirochætes were found by Wise in cases of the destructive local lesion called ulcerative granuloma of the pudenda and have been described by Cleland and

FIG. 90.—Sp. aboriginalis and bacteria. (Bosanquet.)

by Bosanquet (Fig. 90). The organisms are about 12μ in length, ranging from forms a few microns only in length to long forms attaining 18 or 20μ. They are irregular in their curls, thus being clearly distinguished from *Sp. pallida*, and from *Sp. pertenuis*, as seen in the tissues. They closely resemble, however, the pictures given by Castellani of *Sp. pertenuis* as found in the secretion of yaws.

The spirochætes exist in the tissues along with bacterial forms, their relationship to which has already been discussed (see page 58).

SPIROCHÆTA INTERROGANS.

(Stimson, 1909.)

Schaudinn and also Novy suggested that yellow fever might be due to the action of a spirochæte, but failed to find such organisms in cases of the disease. Stimson has recently, by Levaditi's method, found in the kidneys of one such patient spirochætes (?) which often

took the form of a note of interrogation and which he has consequently named *Sp. interrogans.* They are 14μ and upward in length by about $\frac{1}{6}\mu$ in thickness, and the wave-length of the curls is $1\frac{1}{2}$ to 2μ. They were situated in the renal cells and in the tubules, but not in the blood-vessels. The discovery has not been confirmed.

VARIOUS DOUBTFUL SPIROCHÆTES AND SPIRILLA.

Adèle Oppenheimer found spirochætes in the mucus of the **alimentary canal of the dog;** some exhibited snake-like movements (lashing or wriggling), while others had only cork-screw movements, the curls in the body remaining fixed.

These may correspond with the spirilla discovered by Bizzozero in the dog; these organisms had three to seven curls, and were 3 to 8μ long; they lay within vacuoles in the epithelial cells. Salomon also found spirilla in the intestines of dogs, cats and rats; these were longer, and he distinguished three forms—a thick form with seven to nine curls, thicker at the middle than at the ends; a long form with fifteen to twenty-four curls, the axis being often bent; and a form about as long as the first, but with only two to five wavy curls. They had terminal flagella and lay in the mucus of the alimentary canal and also in vacuoles in the cells. Rigaud found spirilla like *Spirochæta pallida* in the stomachs of dogs and cats, lying at the bottom of the peptic glands.[1]

Spirilla were found by Kowalski in the **dejecta of cholera patients,** resembling *Sp. dentium.* They had two to three curls and pointed ends, and did not grow in culture media (Abel). Rechtsamer found that they

[1] *Cf.* Balfour's discovery of spirochætes in ulcers of the intestines of dogs and monkeys, recorded on page 44.

lived a little while in broth: he also noted that the more cholera-vibrios there were present, the fewer were the spirilla, and *vice versa.*

Le Dantec found spirilla in the stools of certain patients suffering from **dysentery.**

Werner found in the **human alimentary canal** two forms of spirochætes—one with long undulations, 4.6 to 7.3μ in length, which may occur in masses (this form is not identical with any of the mouth-spirochætes); and a second, with finer curls, 3.5 to 6.1μ in length, resembling *Sp. dentium.* The former he calls *Sp. eurogyrata* and the latter, *Sp. stenogyrata.*

Smith (Theobald) found spirilla along with comma bacilli (vibrios) in the **intestines of pigs;** they had two to three curls, with a wave-length of 2μ. He could not cultivate them.

Novy and Knapp found spirilla (*Spirillum glossinæ*) in the **stomach of the tse-tse fly;** short forms measured 8μ in length and long ones 15μ. The spirilla were shorter, narrower, and more closely curled than *Sp. obermeieri.* They multiplied by transverse division, the long forms probably being about to divide (see also page 68).

Mezinescu found spirilla in the pus from a case of **calculous pyelitis;** they were from 3.6 to 8μ long, rarely attaining a length of 10 or 12μ. Doerr found spirilla in the pleural and pericardial fluid of a syphilitic subject and thinks they may have been the same as those described by Mezinescu.

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